The Journal of Parasitology

Volume XIV DECEMBER, 1927 Number 2

THE RELATED EFFECTS OF A PARASITE ON A FISH

A RETARDATION OF EARLY GROWTH, THE RETENTION OF LARVAL CHARACTERS AND AN INCREASE IN THE NUMBER OF SCALES

CARL L. HUBBS
Museum of Zoology, University of Michigan

This contribution belongs to a series (Hubbs, 1926, and papers there cited) dealing with the environmental control of the characters of fishes. From these studies I have concluded that modifications of the developmental metabolism induce not only alterations in the actual rate of development, but also secondarily bring about certain definite structural changes. These general conclusions are again confirmed by the results of the present study.

The material used in this investigation consists of a large number of specimens of a cyprinid fish, *Platygobio gracilis,* which I collected with the help of Leonard P. Schultz in a small tributary of the Red River (of the Canadian system), near Springer, New Mexico, on September 21, 1926. These specimens are identified as *P. gracilis* on the assumption that *P. physignathus* is specifically referable to *gracilis;* this point will be considered in a forthcoming report on geographical variation in Platygobio. The species has not before been recorded so far to the southwestward, but we secured other series in the Canadian system of New Mexico, and also in the upper tributaries of the Pecos, in the same state.

Some of the young specimens in the Springer collection, to be exact 68 out of a total of 391, or 17 per cent, differ notably from the others, so much so that on first study I thought that they must represent a distinct species. These aberrant examples furthermore resemble the young of no other series of this species which I have ever seen. They are uniquely abnormal, and appear to represent a new teratological type. They are pale in color and soft in general consistency. Their fin-rays are weak, but the anterior rudimentary rays of the vertical fins are longer than in normal examples. Most of them are pot-bellied and pop-eyed. Their snouts are little produced and their mouths are reduced in size and are more narrowly U-shaped than is usually the case; their barbels are absent or rudimentary; their nostrils frequently joined together on each side; their gill-membranes sometimes more or less free from the

isthmus, and their lateral lines often rudimentary or absent. Furthermore, their scales are generally reduced in size and increased in number, sometimes very markedly so—a notable modification.

Considering this whole array of characters, it is usually an easy matter to decide whether a given specimen in this series is normal or abnormal. The degree of abnormality varies widely, however. The variation is irregular; thus a specimen may be extreme in any one of the aberrant features but nearly normal in others. Each of the characters, considered by itself, grades completely into the condition normal for the species. On account of this observed fact, I was soon forced to the opinion that the aberrant specimens must be referred to *Platygobio gracilis*.

The abnormal features were found to be exhibited only by the young of the year, although a rather large number (94) of yearlings and adults were caught at the same place. Obviously, the abnormality has a lethal correlation.

THE PARASITIC INFESTATION

The population of *Platygobio gracilis* inhabiting this little creek near Springer, New Mexico, is subject to an excessively heavy infestation of internal parasites.[1] The visceral organs, especially the gonads, liver and mesenteries, are often riddled with hundreds or even thousands of trematodes, encysted or preparing to encyst (none were found inside the gut). The trematodes belong to several species; as many as six types seemed to be distinguishable on superficial examination. Cestodes of the genus Proteocephalus, of a species with four suckers, frequently adhere to the inner wall of the gut. A few nematodes are also present in the intestine, and one encysted cestode was found.

The parasites are of course more abundant in the older fishes than in the young. It is a surprising fact, however, that even some of the very small young fishes are already infested with these worms. The occurrence of parasites in the infant fish may be in part attributed to these three factors: (1) The unusually heavy and universal infestation of the older fishes; (2) the occurrence of the young in and about a relatively large and deep weedy pool in the course of this little creek; (3) the food of the young fishes, consisting almost entirely of crustaceans (small ostracods and cladocerans to the exclusion of all else but an occasional larval or adult insect, etc).

A parasite census (Table 1) was made of thirteen normal and of thirteen abnormal young. Examples of the two types were counted in approximate alternation, in order to avoid any cumulative error due to increased skill in locating the worms. A rigid scrutiny for parasites, lasting for about half an hour, was made of the internal organs of each

1. I am greatly indebted to Dr. George R. LaRue for advice relative to the parasitological interpretations made in this paper and regarding the technique of the parasite census, and also for the use of his laboratory and equipment.

specimen. The abdomen was cut open by a U-shaped incision extending back to behind the anus. The viscera were then all removed to a watch glass, and the peritoneal wall was examined (always negatively). The organs one by one were teased apart under a fairly high power, and all parasites removed and counted. It is quite probable that a few very minute trematodes were not found, especially those encysted in the liver; it is highly improbable that a single attached Proteocephalus escaped notice.

TABLE 1.—*Census of Parasites in the Young of Platygobio gracilis*

Material collected in a creek near Springer, New Mexico, on September 21, 1926. Twenty-six individuals were examined, half normal and half abnormal. Each horizontal line gives the number of each type of parasite in a normal or in an abnormal fish of the given length to caudal fin, or in both a normal and an abnormal fish.

Length, Mm.	Total Parasites		Proteocephalus		Trematodes	
	Normal	Abnormal	Normal	Abnormal	Normal	Abnormal
16	..	3	..	3	..	0
17	0	2	0	2	0	0
18	..	3	..	2	..	1
19	0	8*	0	6	0	1
19	1	8	0	8	1	0
20	0	..	0	..	0	..
20	0	..	0	..	0	..
20	2	..	0	..	2	..
21	0	6	0	4	0	2
21	2	6	2	5	0	1
21	..	12	..	9	..	3
22	2*	..	0	..	1	..
23	..	5	..	1	..	4
24	3	..	0	..	5	..
24	8	..	3	..	3	..
25	..	8†	..	5	..	2
25	..	9	..	2	..	7
25	..	10	..	1	..	9
25	..	3	..	2	..	1
26	0	..	0	..	0	..
30	4	..	0	..	4	..
16–23	7	53	2	40	4	12
24–30	15	30	3	10	12	19
Totals	22	83	5	50	16	31

* Including one nematode.
† Including one encysted cestode.

The abnormal young are very much more heavily infested than normal examples of like size (Table 1). This is especially true of the Proteocephalus. Of the thirteen abnormal specimens critically studied for parasites not one was wholly free of this intestinal worm. The number per abnormal fish ranged from one to nine, with an average of nearly four. These aberrant fishes were infested with this worm even though only 16 to 19 mm. long to caudal. Of the thirteen *normal* examples similarly examined, only two (of 21 and 24 mm.) contained Proteocephalus larvae. One of these fishes had two and the other three of these worms. For all the thirteen normal young specimens, the average infestation was less than half a worm per fish, just one-tenth

that of the abnormal fishes. In the six normal fishes 17 to 20 mm. long not one tape-worm was found, whereas the five abnormal individuals 16 to 19 mm. long contained twenty-one of this cestode.

The trematode infestation of the abnormal young was also found to be heavier than that of those which are of normal appearance, but the difference in this case is much less striking. The observed average infestation per fish with Trematoda is only about twice as high for abnormal as for normal fishes. Only one encysted cestode was found (in an abnormal fish), and only two nematodes, one in an abnormal fish and one in a normal one.

The difference in the degree of infestation of the abnormal and normal young fishes was found to be greater at the smaller than at the larger sizes. This point is summarized in Table 2. For the total parasites, these figures indicate that the abnormal young are nearly eight

TABLE 2.—*Relative Infestation of the Smaller and the Larger Young, Normal and Abnormal, of Platygobio gracilis*

Derived from the data given in table 1. The figures indicate the average number of each type of parasite per fish.

Length, Mm.	Total Parasites		Proteocephalus		Trematodes	
	Normal	Abnormal	Normal	Abnormal	Normal	Abnormal
16–23	0.8	5.9	0.2	4.4	0.4	1.3
24–30	3.7	7.5	0.8	2.5	3.0	4.7

times as heavily infested as the normal ones at the smaller sizes, but only twice as badly afflicted at the larger sizes. For Proteocephalus, the aberrant individuals are twenty times as wormy as the normal ones at lengths of 16 to 23 mm., but only about three times as affected at the sizes of 24 to 30 mm. In the case of the trematodes, the difference is less striking; those which are abnormal have on the average three times as many of these worms as the normal young fishes, when smaller, but only about half as many more when larger. Nematodes and encysted cestodes occur too infrequently to yield averages of any significance.

From these and other data I am forced to conclude that the parasitism of these very young fishes by Proteocephalus is by far the most plausible cause of their being abnormal. It is quite probable that the occurrence of trematodes is a contributing factor, especially in those cases in which the infestation with Proteocephalus is light and that with trematodes relatively heavy. Examples of such cases are those of the abnormal specimen 23 mm. long and one of those 25 mm. long, as listed in Table 1.

The fact that the young of Platygobio feed in the locality under consideration almost solely on very small ostracod and cladoceran

crustaceans explains the means of ingress of the proteocephalid tapeworms. Because this is a large-mouthed minnow, it doubtless takes in these crustaceans almost as early as it begins to feed, probably even before the yolk sac is completely absorbed. That the infestation of the

TABLE 3.—*Frequency Table Showing Relative Sizes of the Normal and Abnormal Young of Platygobio gracilis*

Material collected near Springer, New Mexico, on September 21. All of the young of the year were measured, to the nearest millimeter, from tip of snout to extreme structural base of caudal fin. The young of the year are easily distinguished from the yearlings, because the two groups, each compact in itself, are separated by a distinct gap in size. The smallest yearling is 48 mm. long. The reason for the bimodality in the distribution of the length of the normal young is not apparent.

Length, Mm.	Normal Specimens	Abnormal Specimens
13	1	..
14	1	..
15	1	..
16	1	1
17	6	2
18	5	2
19	15	2
20	16	3
21	12	5
22	4	9
23	3	8
24	3	12
25	15	13
26	22	4
27	16	3
28	39	3
29	26	1
30	30	..
31	38	..
32	20	..
33	25	..
34	9	..
35	7	..
36	7	..
37	3	..
Total	323	68
Average size	27.9	23.3

young fishes took place very early in their development is indicated by the facts that the Proteocephalus larvae have already attained a large scolex and have begun to elongate, and that the trematodes have already become mostly encysted, in fishes only 16 to 20 mm. long to caudal. This development of the parasites may not have taken very long, but the growth of the fishes themselves must have been fairly rapid: minnows grow during their first season at the rate of about 0.45 to 0.7 mm. per day (Hubbs, 1921: 273, and 1924: 210). Furthermore, an early infestation would be required to explain the retention of larval characters.

THE EFFECTS ON THE FISH

A very obvious effect of the parasites on these young minnows has been a retardation in the early growth rate (Table 3). The affected fishes average smaller than the normal ones: 23.2 mm. as opposed to 27.7 mm. That the infested fishes are not smaller merely because hatched and exposed to the parasites later in the season is indicated by the fact that the older fishes, all normal, are all very heavily parasitized: so heavy an infection was almost certainly gradually attained during the summer.

These aberrant young of Platygobio obviously appear abnormal because of the retention of larval characters (see Fig. 1). These late embryonic features have been retained as such because their differentia-

Text figure. Normal (above) and abnormal (below) young of *Platygobis gracilis*. Specimens of about the same size; drawn with the aid of a camera lucida. In the abnormal specimen note the pale coloration (the dots represent pigment only); the weak appearance; the short and feeble pectoral fin; the flabby abdomen; the large eye; the united nostrils; the small mouth, and the absence of the barbel.

tion has never been completed. The pale color of these young fishes appears to be due to the poor development of melanophores. Their soft consistency of body and weakness of fin seem to be the result of an incomplete differentiation of muscle and fin-rays. The less-than-usual difference in length between the anteriormost or "rudimentary" dorsal and anal rays and the fully developed rays which follow them in each of these fins represents another incompleteness of development. The fins themselves in these abnormal young, especially the pectorals, have usually not developed to the size attained in normal young of the same length (Table 4)—a condition representing also a decreased differentiation, for the fins in the very early stages grow at a decidedly more rapid

proportionate rate than does the whole body. The pot-bellied condition of the abnormal young is probably to be traced more to a slackened condition of the abdominal wall after the absorption of the yolk sac than to the actual presence of parasites, since the very small normal young have a somewhat flabby and enlarged abdomen, but the larger ones have the abdomen firm and rounded off in line with the rest of the body. The large size of the eye in the abnormal specimens (Table 4) represents yet another retention of a larval feature. So also the lesser production of the snout; the small size and narrowness of the mouth; the poor development of barbels (these are usually absent in the smaller of the abnormal fishes and rudimentary in the larger, although

TABLE 4.—*Proportionate Size of the Eye and Fins in Normal and Abnormal Specimens of Platygobio gracilis*

The size of the eye (measured across the cornea, from wall to wall of the sclerotic cartilage) is expressed in hundredths of the length of the head. The length of the fins (measured from extreme origin to farthest tip, when the fin is depressed) is expressed in hundredths of the standard length to caudal base. The proportions are computed from measurements made in tenths of millimeters. The sixty specimens measured are half normal and half abnormal; the normal ones are 18.3 to 30.6 mm. long, averaging 24.4 mm.; the abnormal ones are 21.5 to 28.7 mm. long, and average 24.9 mm.

	Proportionate Size of Part																	
	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	Averages
Eye																		
Normal							4	1	10	11	3	1						22.4
Abnormal									1	3	6	6	9	2	2		1	25.3
Dorsal Fin																		
Normal								1	6	3	11	7	1			1		23.9
Abnormal							6	5	9	5	2	2			1			21.1
Pectoral Fin																		
Normal						1			6	10	7	2	2	2				22.5
Abnormal	1	4	5	11	3		2	2	1	1								17.4

well developed in all the normal young at hand); the union of the two nostrils, on one or both sides, by the incomplete development of the internarial flap (the flap is actually interrupted in about one-third of the abnormal specimens, and variously weakened in many of the others); the occasional more or less complete separation of the gill-membranes from the isthmus; the frequently rudimentary condition or even absence of the lateral line at sizes at which it is normally developed in the ordinary young, and the absence of scales over the whole body in the smaller abnormal young and on the nape in the larger—all these abnormal conditions represent the retention of larval features, retained because of the retarded differentiation. Great indeed are the structural modifications which this early infestation with parasites has induced in these young fishes.

Even more interesting, from certain standpoints, is the modification in scale numbers which has resulted from the parasitism of these young minnows (Table 5). In this respect as in others some of the abnormal individuals do not deviate from the usual condition. The average num-

TABLE 5.—*Frequency Table Giving the Number of Scales and of Vertebrae in Three Samples of Platygobio gracilis*

The scales are counted as transverse rows, from the shoulder girdle to the structural caudal base. The vertebrae as counted include the hypural plate as one vertebra; the first vertebra is disc-like, the second and third are counted as having their centra completely fused (as an element in the Weberian apparatus); the fourth centrum is nearly normal.

	Scales			Vertebrae		
	Near Normal	Springer, N. M., Abnormal	Powder River, Mont.	Near Normal	Springer, N. M., Abnormal	Powder River, Mont.
40	..	..	..	1	2	..
41	2	..	..	8	7	..
42	2	..	..	3	5	..
43	2	1	..	..	..	1
44	4	..	..	..	..	8
45	6	..	..	..	..	7
46	7	3	..	..	..	1
47	1	1	..	..	..	..
48	2	1	2	..	..	..
49	..	3	3	..	..	..
50	..	..	6	..	..	..
51	..	1	8	..	..	..
52	..	..	4	..	..	..
53	..	..	4	..	..	..
54	..	2	2	..	..	..
55	..	3	..	..	..	..
56	..	..	..	..	..	..
57	..	1	..	..	..	..
58	..	1	..	..	..	..
59	..	2	..	..	..	..
60	..	3	..	..	..	..
61	..	2	..	..	..	..
62	..	..	..	..	..	..
63	..	..	..	..	..	..
64	..	..	..	..	..	..
65	..	2	..	..	..	..
Totals	26	26	29	12	14	17
Averages	44.7	54.3	51.0	41.2	41.2	44.5

ber of scales, however, is about ten higher than in the normal young (54.3 as contrasted with 44.7), and the number is greatly increased in extreme cases, as the table shows. The wide dispersion in the scale counts for the parasitized specimens is indeed remarkable.

Very significantly, the vertebrae do not show a like increase in the abnormal specimens. The number of vertebrae and scales is nearly the same in most minnows, and in their mutual variation they exhibit, as I will show in a subsequent paper, a high degree of positive correlation: an increase in the scale number is usually associated with an increase in the number of vertebrae. This correlation is racial as well as individual. Thus I find that *Platygobio gracilis* in Montana shows a somewhat

similar increase in the number of vertebrae and of scales as compared with the normal examples of the New Mexico race under treatment (Table 5). Further data on the geographical variation in the number of scales and vertebrae in *Platygobio gracilis* are being gathered for a subsequent paper. The increase in scale number in the abnormal parasitized young from New Mexico is a striking example of disharmonious variation, but is readily explained on the assumption that the abnormality of these young is caused by the parasites, which cannot enter to effect this change until after the time when the number of somites and hence of vertebrae is fixed.

DISCUSSION

So far as I know, this is the first time that parasites have been suspected of so greatly modifying the specific characters of a host fish. The effect has been such as to lead one very naturally to the erroneous conclusion that the examples which have been molded into a new appearance by the early infestation are representative of a distinct species. This source of individual modification can not well be ignored by ichthyologists. To even a greater degree should these results be considered by the student of geographical speciation. Even an occasional modification of characters by parasites in one region but not in another might produce differences in proportions or scale number that would measure up to the criterion of statistical significance.

The apparently new teratological type exemplified by these parasite-modified fishes indicates the desirability of considering parasites as a possible cause of other terata. I strongly suspect that certain modifications of proportions and even of coloration may be induced by parasites. This suspicion is especially warranted in the case of the microphthalmic condition, which has occasionally a high local incidence. The rare occurrence of fishes with an abnormally high scale number may perhaps find a general explanation as an effect of an early infestation with parasites. The parallel effects of early infestation of parasites with other conditions retarding development (Hubbs, 1924, 1926, etc.) is remarkable. The protraction of the larval-postlarval differentiation into a stage of general development or condition of tissues in which its completion is rendered impossible, despite the fact that the differentiation would on general theory be expected to proceed under these conditions at least as far as under accelerated circumstances of development, is very well illustrated in the effects of the parasites on these young fishes.

The remarkable increase in the number of scales exhibited by these young fishes early infested with parasites falls likewise into line with the general effects of conditions retarding the early development. The reason for this increase in the number of scales under slackened develop-

ment appears to be as follows. Under such conditions the scales do not form as early as in normal or accelerated young; this is strikingly true in the present instance as well as in others in which the cause of the protracted development is different. But when they do differentiate, the scales are laid down at roughly the same focal size and at approximately the same absolute distance from one another as in the other case. Since the body base is larger, the number of scales is increased.

The foregoing observations and interpretations naturally suggest further investigations in the same field. In particular, the experimental infestation of very young fishes in an effort to produce teratological effects, and to analyze the physiological effects involved, should be undertaken. It might be possible in this way to shed light on the intricate problem of the mechanism of development and of speciation.

Literature Cited

Hubbs, Carl L. 1921.—An ecological study of the life-history of the fresh-water atherine fish Labidesthes sicculus. Ecology, 2:262-276.

1924.—Seasonal variation in the number of vertebrae of fishes. Pap. Mich. Acad. Sci., Arts & Letters, 2:207-214.

1926.—The structural consequences of modifications of the developmental rate in fishes, considered in reference to certain problems of evolution. Amer. Nat., 60:57-81.

HERPETOMONAS ARCTOCORIXAE SP. NOV. ENTOZOIC IN THE INTESTINE OF THE WATER-BOATMAN *ARCTOCORIXA INTERRUPTA* SAY

ELERY R. BECKER
Iowa State College

A number of species of aquatic Heteroptera from the vicinity of Baltimore, Md., and Cold Spring Harbor, L. I., were examined for intestinal flagellates by the writer during the years 1921 and 1922. At that time *Crithidia gerridis* Patton 1908 was reported from the gut of *Gerris remigis, G. marginatus, G. rufoscutellatus,* and *Microvelia americana* (see Becker 1923). During the same period flagellates of the genus Herpetomonas were found in the intestine of the water-boatman, *Arctocorixa interrupta* Say, common in the vicinity of Baltimore. Permanent slides of these flagellates were prepared at that time by the iron hematoxylin method. These slides furnished the material upon which the present study was made. The writer wishes to take this opportunity to thank Dr. H. B. Hungerford of the University of Kansas for identifying the insect host.

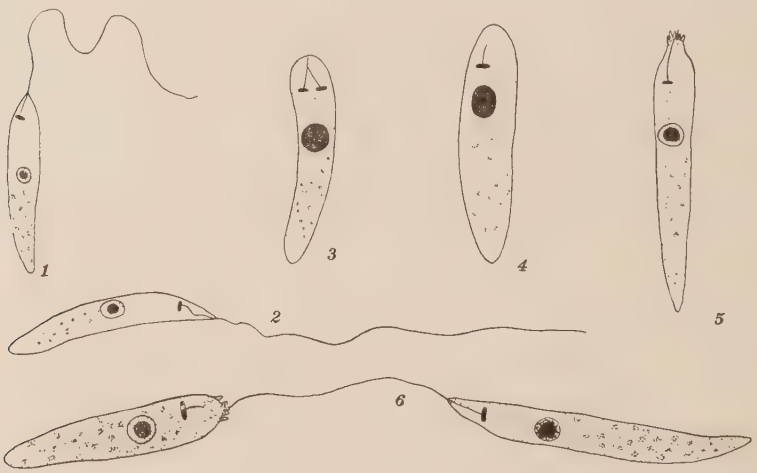
Both nymphs and adults were found infected with herpetomonad flagellates, but only to the extent of about five per cent. The infections usually were very light, the only heavily infected individual being one small nymph. The lightness of the infections leads one to suspect that perhaps they are only secondarily acquired by these corixids, and that they are primarily infections of the intestines of other insects which have fallen prey to these predaceous forms. This type of flagellate, however, was never encountered in any other of the many insects which inhabit the water or water's edge.

Two principal types of the flagellate were encountered, the flagellated and the non-flagellated (Text fig.). That both types are viable organisms is attested to by the active movements of the former, and the dividing forms of the latter (3). I propose *Herpetomonas arctocorixae* as the name of this flagellate.

The general structure of the flagellated form is similar in a general way to that of *Herpetomonas* (*Leptomonas*) *ctenocephali, H. pulicis,* and *H. jaculum as contrasted with H. muscae-domesticae.* It will be remembered that the latter flagellate has a predominant bi-flagellated form (owing to precocious division), prominent marginal granules at the points where the flagella leave the body (Becker 1923 a), and a large parabasal body often heart-shaped when the division of the parabasal body has been initiated. The body, not including its flagellum, varies

from 10 to 19μ in length, and at the level of the nucleus is about 2μ in width. The flagella measure from 17 to 23μ in length from the point where they leave the body to the free tip.

The cytoplasm usually stains densely. The nucleus is of the vesicular type, with usually one large central karyosome. The parabasal body is rod-shaped, and lies at right angles to the long axis of the body. It stains green with Janus green B, after the fashion of the parabasal bodies of *Trypanosoma lewisi* (see Shipley 1916), *Crithidia gerridis* (see Becker 1923a) and *Herpetomonas muscae-domesticae* (see Becker 1923b). Causey (1925) and Grassé (1926) have confirmed these observations; the former in the case of the parabasal body of the herpetomonad form of *Leishmania brasiliensis,* and the latter in *Herpetomonas jaculum* and



Text fig. 1, 2, flagellated types; 3, 4, nonflagellated or gregariniform types, former with a divided parabasal body; 5, nonflagellated type with cuticular bristles at anterior end; 6, association of flagellated and nonflagellated individuals.

H. pyrrhocoris. The basal granule from which the flagellum takes its origin is not visible as in *Herpetomonas muscae-domesticae.* The condition is similar to that in *Crithidia gerridis* where the flagellum seems to arise directly from the parabasal body. In such cases the basal granule probably either lies within the parabasal or is closely adpressed against its anterior surface. The flagellum is single and extends anteriorly from the body.

The non-flagellated (or gregariniform) individuals are markedly larger than the majority of the flagellated forms, both in respect to width and length (Text fig. 3 and 4). The nucleus of these forms is usually considerably enlarged, as compared with the flagellated forms, and stains more or less homogeneously. The parabasal body gives rise to the basal intracytoplasmic portion of the flagellum which terminates

either at (3) or before (4) it reaches the margin of the body. These forms are not motile. A number of agglomeration rosettes were encountered, formed by a congregation of thirty to fifty about a mass of intestinal débris with their anterior ends directed inwards. They resemble the rosettes of Paramecium feeding upon a mass of bacteria or collected about a small gas bubble.

A number of nonflagellated individuals which may be transitional forms were seen. These differ from the others in that the cuticle at the anterior end is frayed out brush-like (5). It appears as if the proximal part of the flagellar sheath had become shredded with the disappearance of the flagellum. A curious association of a flagellated and a non-flagellated individual was seen a number of times (6). The tip of the flagellum of one member of the pair is attached to the point where the flagellum of the other individual formerly left the cell. Despite violent lashings of the flagellum the union was not broken. It is doubtful if there is any broad significance in such associations, for it is well known that herpetomonads have the ability to adhere with the tip of their flagellum to the cells of the intestinal epithelium. But just why the contact was always at this particular point and no other is hard to understand, unless perchance the non-flagellated cell is stickier in this region, and any chance contact by the tip of the flagellum of another individual would unite them.

SUMMARY

Herpetomonas aretocorixae sp. nov. is an inhabitant of the gut of the water-boatman, *Arctocorixa interrupta*. It occurs in both the flagellated and non-flagellated or gregariniform forms. The infections are usually rather light, suggesting the possibility that the infection is secondarily acquired from some insect serving as prey to this predaceous insect. A curious association of flagellated and non-flagellated forms is reported. In such cases the tip of the flagellum of one individual is attached to the point in the other where the flagellum formerly existed.

LITERATURE CITED

Becker, E. R. 1923a.—Observations on the Morphology and Life Cycle of *Crithidia gerridis* Patton in the Water-Strider, *Gerris remigis* Say. Jour. Paras., 9:141-152.

1923b.—Observations on the Morphology and Life History of *Herpetomonas muscae-domesticae* in North American Muscoid Flies. Jour. Paras., 9:199-213.

Causey, David. 1925.—Mitochondria in *Leishmania brasiliensis* Vianna, 1911. Univ. Cal. Publ. Zool., 28:19-28.

Grassé, Pierre P. 1926.—Contribution a l'étude des flagellés parasites. Arc. Zool. Exper., 65:345-602, 12 pl.

Shipley, P. G. 1916.—The vital staining of mitochondria in *Trypanosoma lewisi* with Janus green. Anat. Rec., 10:439-445.

THE EGG PRODUCTION OF *ASCARIS LUMBRICOIDES* *

H. W. Brown
AND
W. W. Cort

The daily egg production for several different species of parastic worms has been estimated by a comparison of the number of worms obtained by treatment with the egg output as determined by the dilution egg count. By this method Stoll (1923) obtained a figure of about 9,000 eggs per day for a female of *Necator americanus* and Sweet (1924) and Cort, Stoll and Grant (1926) estimated the daily output of a female of *Ancylostoma duodenale* to be several times as large. For *Fasciolopsis buski* Stoll, Cort and Kwei (1927) have given an estimate of a daily output of about 25,000 eggs per worm. Faust and Khaw (1926) reported that the number of eggs produced by *Clonorchis sinensis* depended on the kind of host. For infestations in the cat their estimate was 2,400 eggs per day per worm, for the guinea pig 1,600, and for the dog 1,100.

An egg output much higher than for any of these species would be expected for *Ascaris lumbricoides,* since these worms are of such large size and contain so many eggs in their uteri. Cram (1925) has recently estimated that the number of eggs in a single female ascaris may be as high as 27,000,000. We were, therefore, to some extent prepared for the high egg production figures obtained for this parasite from the study of two cases in which one of us (Brown) was able to make an egg-worm correlation in Panama.

The first case was a boy 5 years of age who was found to be passing in the total fecal output for a single day approximately 10,064,000 ascaris eggs. After treatment 34 male and 43 female ascarids were obtained which gave about 234,000 eggs per day for each female. The average length of the female worms from this case was 24 cm. and their average estimated volume was 4.6 cc. Since egg counts were made for only one day before treatment the figure obtained in this case must be considered as only tentative.

The second case was a 13 year old boy who was shown by egg count to have passed about 735,000 unfertilized ascaris eggs in a three day period. After treatment a single large female containing only unfertil-

* This paper is a contribution from the Department of Helminthology of the School of Hygiene and Public Health of the Johns Hopkins University. The work was carried out with the cooperation of the International Health Division of the Rockefeller Foundation.

ized eggs was passed, which gives a daily egg production of about 245,000 eggs. This worm was of large size having a length of 27.3 cm. and a volume of about 5.2 cc. In these two cases the eggs per gram per female worm averaged close to 2,000. We present this figure as a tentative egg-worm ratio for the human ascaris in the hope that others, who are favorably situated for such work, will check our findings with larger numbers of cases.

It is possible, however, to go a step further and to determine whether the application of this figure to series of egg counts of ascaris gives results in agreement with the general information available on the size of infestations with this parasite. We have available from the examinations of the China Hookworm Commission and the Hookworm Expedition to Panama two very large series of unpublished egg counts for *Ascaris lumbricoides*. If we postulate that when large numbers are considered there will be approximately an equal number of males and females in this species we can use 1,000 eggs per gram per adult worm in estimating the number of worms which correspond to our egg count figures. We know further that there are almost always present in ascaris infestations different sizes of worms too young to produce eggs which would have to be added to give the true worm burden of an individual or a group. Since it would be very difficult to estimate the percentage of such young forms, we shall make our comparisons only in numbers of adult worms present. The estimation of the numbers of worms present in different individuals or in groups by applying the figure of 1,000 eggs per gram per adult worm to our egg counts gives infestations of sizes which appear quite reasonable in the light of what we know of ascaris infestations from counts of worms after treatment. While we must consider this number as only a rough approximation when applied in this way it becomes immediately evident that any very considerable reduction of it would place the worm burdens of individuals and groups at an unreasonably high figure. It would be anticipating too far the accounts which will be published later to give here any considerable number of the individual egg counts or the averages for any large series of groups. It seems worth while, however, to give a few examples of the results obtained in numbers of worms when the factor of 1,000 is applied.

Our Panama series contains counts for ascaris on 2,247 individuals from twelve different population groups. Thirty-two of these cases had counts of over 200,000 eggs per gram, with an average count of about 345,000. If we use the factor of 1,000 eggs per gram per adult worm, we obtain an average infestation for this highest group of 345 adult ascarids, plus whatever immature forms were present. A group of 86 children under 15 years of age from one of the poorer sections of a city in Panama had egg counts for *Ascaris lumbricoides* varying from 1,800 to 761,200 per gram of feces, with an average of about 72,000.

The estimated number of *adult* worms in this group would then vary from 1 or 2 to 760, and the average would be about 72 per individual. In a group of 51 school children from 10 to 14 years of age examined near Canton, China, the average egg count for ascaris was 50,200, which would give an average of 50 adult ascarids. It seems evident to us that the average egg production of Ascaris cannot be very much lower than in the two cases studied without making the estimate of worm burdens in such individuals and groups as those given above unreasonably high. Even allowing for great variability, these figures show an extreme fecundity and demonstrate that in the human ascaris, even with its eggs so unusually resistant to environmental conditions, and without the hazards of free larval stages or life in an intermediate host, there is an almost unbelievable reproductive wastage.

Literature Cited

Cort, W. W., Stoll, N. R., and Grant, J. B. 1926.—Researches on Hookworm in China. I. Problems and methods of attack. Amer. Jour. Hyg., Mon. Ser., No. 7, pp. 1-32.

Cram, E. B. 1925.—The egg-producing capacity of *Ascaris lumbricoides*. Jour. Ag. Res. 30:977-983.

Faust, E. C. and Khaw, O. 1926.—The egg-laying capacity of *Clonorchis sinensis*. Proc. Soc. Exp. Biol. and Med., 22:606-607.

Stoll, N. R., Cort, W. W. and Kwei, W. S. 1927.—Egg-worm correlations in On the relation between the number of eggs found in human feces and the number of hookworms in the host. Amer. Jour. Hyg. 3:156-179.

Stoll, N. R., Cort, W. W. and Kwei, W. S. 1927.—Egg-worm correlations in cases of *Fasciolopsis buski*. Journ. Parasit., 13:166-172.

Sweet, W. C. 1924.—The effect of carbon tetrachloride on the egg laying powers of the female hookworm. Amer. Jour. Hyg., 4:691-698.

DRACUNCULUS GLOBOCEPHALUS N. SP., FROM *CHELYDRA SERPENTINA* *

J. G. Mackin

In the fall of 1926 and the spring of 1927, a number of a new species of nematode were removed from the mesenteries and body cavity of the snapping turtle, *Chelydra serpentina*. The parasites are of particular interest in view of their close relationship with the famous human parasite, *Dracunculus medinensis*. Collection records include records from Ada City Lake, Ada, Oklahoma; a small limestone sink lake near Alto Pass, Illinois; and an oxbow pond near Urbana, Ill. It is interesting that several specimens of *Chelydra serpentina* from running streams did not harbor the parasites. The nematodes were found actively moving in the mesenteries of the hosts, usually in the posterior region dorsal to the large intestine. Some few were located in the more anterior regions.

DESCRIPTION OF ADULT

The body is long and threadlike, with not much difference in diameter throughout the entire length. The posterior region is rather attenuated and ends in a small conical spike in both sexes. A slight constriction near the head gives the appearance of a neck. The anterior region is bluntly rounded and without lips. There are eight cephalic papillae symmetrically arranged around the mouth (Fig. 3). Situated dorsoventrally is the largest pair, which projects anteriad. Next in size is the lateral pair, each one of which is double, having two nerve endings to each of the cone-shaped papillae. Four submedians complete the list. About 0.9 mm. from the anterior extremity is a single pair of cervical papillae (Fig. 5), situated on the lateral lines. The lateral fields are broad; from one eighth to one sixth of the total bodily circumference. They are inconspicuous in toto mounts. The median lines are very much reduced.

The males are very small compared with the females, ranging from 16 to 20 mm. in length by 170 to 220μ in thickness. The caudal papillae of the male consist of a single minute preanal, and one pair of dome shaped postanals, situated ventro-laterally, one slightly anterior to the other. The tail of the male in fixed specimens is coiled in a spiral of from one to four loops (Fig. 7). The females vary greatly in length. The smallest found measured only 30 mm. and the largest 133 mm. The thickness varies from 280 to 680μ. Apparently the females increase in size as the embryos develop.

* Contributions from the Zoological Laboratory of the University of Illinois, under the direction of Henry B. Ward, No. 308.

The male genital system consists of a single straight tube beginning with the testis one fourth of the body length from the anterior end, and running posteriad to the cloaca. The testis merges into the vas deferens almost imperceptibly. The latter organ near the cloaca fills nearly the whole of the body cavity, then tapers rapidly to its junction with the alimentary canal. The right spicule has the form of a long narrow needle without wings, 0.8 mm. long and 5μ thick. This spicule was always found to be extruded from the genital cone more than one third of its length. The left spicule is 0.2 mm. long and 10μ thick. The proximal third is a hollow tube flaring at the end; the distal two thirds has the form of two tubes joined laterally, the left side continuous with the proximal end and the right offset. The two merge distally into a fine point. This spicule was found extruded in most specimens. A number of single celled glands lie posterior and lateral to the cloaca, with the ducts emptying into the latter organ very close to the anal opening.

Two short ovaries are present in the female, one at each end of the body. The anterior one is wound around the glandular portion of the esophagus and extends forward to within 2 mm. of the anterior extremity of the body (Fig. 1). It is telescoped slightly into the larger uterus at the junction of the two, about 4 mm. from the head. The posterior ovary is wound around the posterior end of the intestine and reaches to the anal region (Fig. 4). There is no rachis present. There is no region that corresponds to a seminal receptacle. Apparently fertilization takes place throughout the uterus as all the embryos in any one individual are in about the same stage of development (Fig. 6). The two branches of the uterus form one long continuous tube joining the two short ovaries. The whole body cavity is filled with the uteri, more or less crowded with the developing embryos (Fig. 2). Theoretically there are two uterine tubes; actually there is only one, for there is no constriction of the two at their junction with the vagina, even in those young individuals in which that organ is not atrophied. The uterine wall is composed of two layers; the outer very thin and the inner nucleated and about 3μ thick.

The vulva is situated a short distance posterior to the center of the body. The opening is very small, communicating with a bulbous structure. This bulb like structure connects with a very small vaginal lumen. The vagina, after running dorsally to the region of the left lateral line, turns forward, lying in the angle formed by the intestine and uterus. It is probable that the entire vagina and vulva atrophy in females with fully formed young, as evidenced by progressive degeneration in less mature specimens (Fig. 8). Of the several females sectioned in the vulvar region, one, the smallest, had the vulva and vagina entire. In a larger specimen degeneration had progressed to the point of rendering

the organs useless. The largest specimens examined had left only a short section of the vagina, attached to the vulva.

The digestive system consists of a short muscular portion of the esophagus, a much longer glandular portion, and the intestine. There is no buccal cavity or pharynx. The mouth is more or less triangular in shape, opening directly into the esophagus. The muscular portion of the esophagus has the typical glandular arrangement; at the junction of the muscular with the glandular portion of the esophagus, the glands begin to enlarge, to the extent that the appearance is of two large sacs attached to the outside. These sacs are hollow for part of their length. Two ducts from the lumen of the esophagus communicate with the glands. A third gland, much larger than the two just described, lies along the opposite side of the esophagus, extending from within 0.5 mm. from the anterior extremity posteriad over a third of the body length. This gland ends at the junction of the posterior esophagus with the intestine. While it could not be demonstrated that the origin of this gland is the same as that of the other two, there is not much doubt that this is true, considering that the strucure of the three is very much alike. All are heavily granular, and possess a single very large nucleus; which in the large gland in one individual reached the enormous length of over 3 mm. The lumen of the posterior esophagus is practically occluded for most of its length. It is represented by a very small slit. The intestine is simple and ends blindly in the mature females. The posterior end is attached to the ventral body wall in the curvature of the tail by a strand of tissue. No anal opening could be found.

A large gland functions as a conveyor of the canals of the excretory system. This gland is attached to the left lateral line in the anterior region of the body. A smaller gland on the right side also carries a branch of the canal. The excretory pore is located from 1 to 1.5 mm. from the anterior extremity, varying with the size of the individual.

A few facts of the life history have been established. It was discovered that the females with living young, when placed in tap water would invariably burst, liberating the myriads of embryos in the water. This is apparently the normal medium for embryos at this stage. When placed in normal salt solution they lived for as much as seven days, but were sluggish for the whole period. Others kept in tap or pond water were active several days after those in normal salt solution had died. Their activity rapidly decreased on the ninth and tenth days and all were dead by the twelfth.

Some of the larvae, shortly after liberation from the parent, were fed in large numbers to several different species of Crustacea, including Ostracoda, Copepoda, and Cladocera. One species of copepod, *Cyclops bicuspidatus,* readily took the wriggling larvae, which promptly bored through the walls of the alimentary tract into the body cavity. None of

the other Crustacea could be infected. Cyclops were not inconvenienced by the presence of the worms except when a very heavy infection occurred, in which cases the alimentary canal became so lacerated as to cause death. It is probable that over infection in nature would rarely or never occur.

Further studies on the anatomy and life history are being made in hopes of clearing up some of the remaining problems and incidentally to throw light on some of the obscurities concerning the genus. In spite of the fact that *Dracunculus medinensis* has probably been known longer than any other parasite, much remains to be done on the anatomy and life history. With the exception of a few minor details of anatomy, size, and primary host, *Dracunculus globocephalus* and *Dracunculus medinensis* are almost identical. It is deemed important that an opportunity is here afforded to study males of the genus. No description of the male Dracunculus has heretofore been given; indeed only two are certainly known to have been found. Leiper (1907) removed these from a monkey which had been artificially infected. He gave no description beyond the length (22 mm.). Neumann (1895) described a new species, *Dracunculus dahomensis* from the python. He found one male of which he gave a short description. Yorke and Maplestone (1926) list this species as a doubtful synonym of *Dracunculus medinensis*.

References Cited

Leiper, R. T. 1907.—The Etiology and Prophylaxis of Dracontiasis. Brit. Med. Jour. 1:129-132.

Neumann, G. 1895.—Sur une Filaire (*Filaria dahomensis* n. sp.) du Python de Natal, voisine de la Filaire de Médine. Bull. Soc. zool. de France, Paris, 20:123-127.

Yorke, W. and Maplestone, P. A. 1926.—The Nematode Parasites of Vertebrates. 536 pp., 306 figs. Philadelphia.

Explanation of Plate

The scale represents 0.1 mm., except in figure 6 in which it is 0.01 mm.

Fig. 1.—Lateral view of the anterior end of the female, showing the large esophageal gland, location of the excretory pore, and anterior end of the ovary.

Fig. 2.—Embryo from the uterus of the female.

Fig. 3.—Face view of the head to show the location of the papillae.

Fig. 4.—Posterior extremity of the female.

Fig. 5.—Anterior extremity of the male, dorsal view, locating the cervical papillae.

Fig. 6.—Developing embryo.

Fig. 7.—Posterior extremity of the male.

Fig. 8.—Stereogram of the vulvar region of the female, showing manner of disintegration of the vagina.

PLATE V

SOME REMARKS IN CONSEQUENCE OF THE RESEARCHES OF SVENSSON AND KESSEL ON NECATOR AND ANCYLOSTOMA LARVAE *

P. H. VAN THIEL

The problem of the differential diagnosis of the strongyloid larvae (filariform stage) of the hookworm of man and of animals is one to which attention must be paid by tropical hygienists all over the world, while it is not possible to seek for larvae of the hookworm of man in polluted soil without a good knowledge of this subject. Also it is desirable that persons who are making researches on this subject be acquainted with the literature hitherto published.

The recent researches from the American side of Cobb, of Svensson and of Svensson and Kessel, have attracted attention, but the publications of Dutch authors seem to have slipped out of sight. For this reason I should like to point out this oversight.

The author (1924) first questioned the opinion of Looss, that there was no differences between the strongyloid larvae of hookworms of man and of the dog, after Schuurmans Stekhoven-Meyer after a very minute, especially biometrical, examination had expressed shortly before, the wish to join themselves to the opinion of Looss. In that publication the author elucidated by microphotographs (1) the difference in the structure of the chitinous part of the buccal cavity of the larvae of Necator and of *Ancylostoma caninum;* (2) the much more pronounced transverse striature on the sheath of the first in comparison to the second; this publication is mentioned by Hall in the JOURNAL OF PARASITOLOGY for 1925, on page 229.

In 1926 the author described more minutely the differences between these larvae and especially the structure of the esophagus, the union of esophagus and intestine and of the tail, discussing the recently printed publications of Cobb and of Svensson. The opinion of Cobb was criticised, that the chitinous buccal armature would function as a spear. Since the publications of Svensson and Kessel another one by Schuurmans Stekhoven (1926) has appeared; in this the observations of the author are affirmed in the main; however, as to the strongyloid larvae of Necator and of *Ancylostoma caninum,* new points of difference are described, and explained by very minute drawings, concerning the excretory apparatus, the presence or absence of lips, the situation of the amphids and of the papillae on the head.

Lately in a meeting of the Dutch Society of Tropical Medicine (to be published in the reports of that meeting in the "Nederlandsch Tijd-

* From the Laboratory of Tropical Hygiene of the Institute of Tropical Medicine at Leyden.

schrift voor Geneeskunde") the author has announced that the larvae of the dog hookworm which were described by him before, appear to be not of *Ancylostoma caninum,* but of *Uncinaria* (*Dochmoides*) *stenocephala.* This worm is known to occur in temperate countries, though very little is recorded on its distribution in the tropics, but the odds are that it will be found there, too. The parasite, which possesses no teeth in the buccal capsule as has been described well by Fülleborn, is easily determined.

The larvae of this species can be recognized without any difficulty. The strongyloid larva has in common with that of *Ancylostoma duodenale* (1) the presence of three lips; (2) the form of the chitinous part of the buccal cavity; (3) the little clear and narrow intestinal lumen. It differs from it by the following characteristics: (1) the irregularity in the structure of the esophagus bulb, which is caused by the form of the esophageal glands situated there, is not visible in Uncinaria without a high power immersion objective; the contrary is true of Ancylostoma; this is not caused by their absence, but by their very delicate character. (2) In Uncinaria the tail of the sheath is strikingly obtuse, in contradistinction to *Ancylostoma duodenale* and Necator where it ends very sharply. Though not a sufficient number (merely sixty) measurements could be made to calculate the coefficient of correlation, it is obvious of the following extreme values, that the distance from the indication of the anus on the sheath to the point of the tail is not the same in Uncinaria as in the other two worms.

	Total Length	Anus-point of the Tail (on the Sheath)
Ancyl. duodenale	672-784 μ	121-136 μ
(Pure culture out of one female)	512-592 μ	95-110 μ
Necator americanus	592-704 μ	110-133 μ
(Once)	512 μ	99 μ
Uncinaria	576-656 μ	87-91 μ

(3) The intestinal valves in Uncinaria are not so obvious as in *Ancylostoma duodenale.*

References Cited

Fülleborn, F. 1924.—Bemerkungen über die Identifikation von "Hakenwürmern." Arch. Schiffs Tropen-hyg., 28:12-15.

Schuurmans Stekhoven, J. H. en Schuurmans Stekhoven-Meyer, A. W. 1924.—Voorbereidende onderzoekingen met betrekking tot de differentiatie van de larven van de mijnwormen van mensch en hond. Tijdschr. v. Vergel. Geneesk., 10, Afl. 2-3.

Schuurmans Stekhoven, J. H. 1926.—De Nematodengeslachten Anchylostoma en Necator. Kon. Acad. Wetens. Amsterdam, 35:1185-1198.

Svensson, R. M. and Kessel, J. 1926.—Morphological differences between Necator and Ancylostoma larvae. Jour. Parasitol., 13:146-153.

Thiel, P. H. van. 1924.—De differentieeldiagnose van de strongyloide larven van *Necator americanus* en *Anchylostoma caninum.* Tijdschr. Vergel. Geneesk., 10, Afl. 4. (transl: Acta Leidensia, I, 1926: 306).

1926.—Verdere onderzoekingen omtrent de differentieeldiagnose van de strongyloide larven van *Necator americanus* en *Anchylostoma caninum.* Geneesk. Tijdschr. Ned. Ind., 66:1-10.

1926.—Diagnostic différentiel des larves strongyloides du *Necator americanus* et de *l'Ankylostoma caninum.* Ann. Par. hum. comp., 4:228-232.

POLAR PLANIMETER MEASUREMENTS OF CYSTS OF *IODAMOEBA WILLIAMSI* *

SEPTIMA C. SMITH

Unlike the cysts of most of the other species of amoeba living in man, those of *Iodamoeba williamsi* are not spherical but irregular in shape. The size of the cyst has been recognized by protozoologists for many years as a criterion of great value in distinguishing between species and races of the parasitic amoebae. Hence it is important to be able to measure those of *I. williamsi* accurately. Heretofore, they have been measured, by the various investigators who have studied the organism, in linear units, that is in terms of length and breadth. But, due to the marked irregularity in shape, all have agreed that it is difficult to give a single measurement of size. Dobell (1919) employed as a measure of the size of this species, the average of the greatest length and the greatest width. Taliaferro and Becker (1922), following this precedent, found that most cysts lie between 6.4 μ and 16.6μ, with an average of 9.1μ in diameter. They also found a daily variation in shape in the same infection. Since the forms are so aberrant and show such fluctuations, I decided that a more accurate unit of measurement would be that of the area. Hence camera lucida drawings of 100 cysts obtained from the same host on two different days about one week apart were sketched and the areas measured with a polar planimeter. All sketches were made with a 12.5 $\times$ ocular, and a 4 mm. objective, which gave a magnification of 2,000 diameters. Preceding the operation, the planimeter was accurately and carefully calibrated with the microscope used, with the idea of interpreting the results in terms of microns. It was found that 1 P P unit $=6{,}450{,}000\mu^2$. This number, divided by the square of the magnification reduced the unit of the drawing to $5.5\mu^2$. Hence the area of the cyst itself is the product of this constant factor and the area of the drawings. The mean of the areas based on linear units is 70.92 ± 1.2, and the standard deviation is 18.49 ± 0.88. The mean of the planimeter areas is 75.36 ± 1.3, and the standard deviation is 18.84 ± 0.90.

Table 1 gives the frequency distribution of the areas of the cysts measured, while text-fig. 1 is a histogram of the frequencies grouped according to certain definite class limits. The diagram gives an adequate picture of the real distribution of the areas in this series. The skewness

* From the Department of Protozoology, School of Hygiene and Public Health, Johns Hopkins University. The writer wishes to express her appreciation to Dr. L. J. Reed, Professor of Biometry and Vital Statistics, School of Hygiene and Public Health, Johns Hopkins University, for advice and criticism in preparing this paper.

TABLE 1.—*Frequency Distribution of Polar Planimeter Areas of 100 Cysts of I. williamsi*

Area Polar Planimeter Units	Area in sq. μ	Frequency	Class Limits for Frequency Distribution Grouped in Limits of 10 μ^2	Frequency
3- 3.9	16.5- 21.9	1	10- 19.9	1
4- 4.9	22.0- 27.4	0		
5- 5.9	27.5- 32.9	0	20- 29.9	0
6- 6.9	33.0- 38.4	1	30- 39.9	3
7- 7.9	38.5- 43.9	3	40- 49.9	5
8- 8.9	44.0- 49.4	2		
9- 9.9	49.5- 54.9	8	50- 59.9	12
10-10.9	55.0- 60.4	6		
11-11.9	60.5- 65.9	8	60- 69.9	16
12-12.9	66.0- 71.4	7		
13-13.9	71.5- 76.9	12	70- 79.9	17
14-14.9	77.0- 82.4	9		
15-15.9	82.5- 87.9	15	80- 89.9	19
16-16.9	88.0- 93.4	13		
17-17.9	93.5- 98.9	3	90- 99.9	18
18-18.9	99.0-104.4	6		
19-19.9	104.5-109.9	4	100-109.9	7
20-20.9	110.0-115.4	1	110-119.9	2
21-21.9	115.5-120.9	1		
Totals.....		100		100

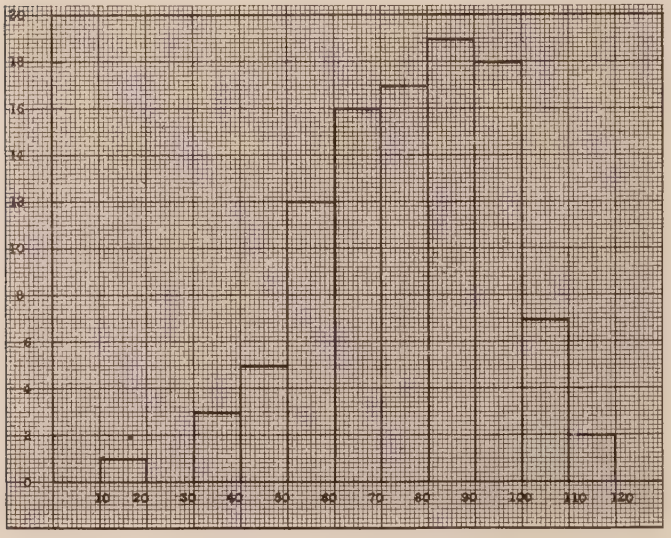


Fig. 1.—Histogram of the frequencies grouped according to certain definite class limits. The vertical line indicates frequency; the horizontal the area in square micra.

of the distribution is apparent, as the greatest number of individuals occurs not at the mid-line of the range, but in the 80-90μ^2 class, whereas the mean area falls, as stated, at 75.36μ^2. It will be noticed that the skewness in the histogram is in the opposite direction from that usually observed in biological studies. The direction which the skewness takes may be due to some simple point in the set-up of the data, or, on the other hand, to some as yet unobserved fundamental biological factor. As planimeter areas are based on two dimensions, they are probably more accurate than linear ones, founded on a single dimension. But a three-dimension measure would, presumably, be more accurate than a planimeter

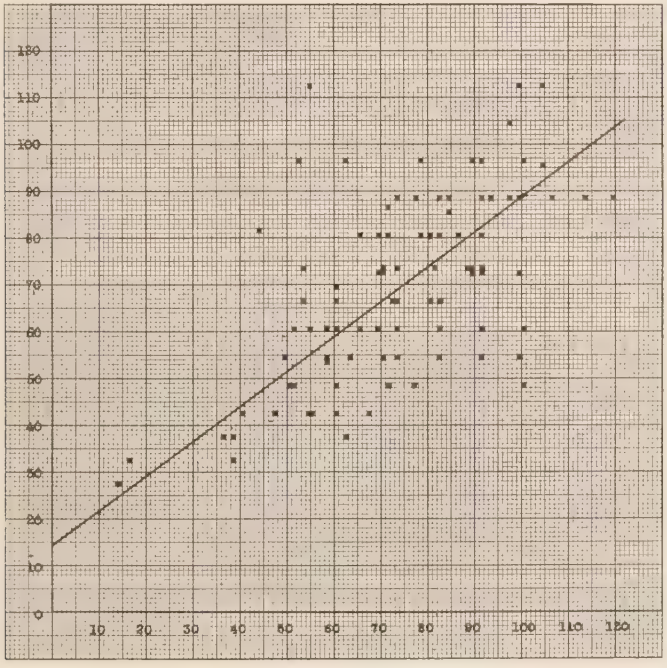


Fig. 2.—Diagram representing variations in areas of 100 cysts based on the two methods used. The vertical line shows areas based on linear units, the horizontal the planimeter areas — both in square micra.

unit, and thus might offer some solution to the problem. The only answer which I can give at present is that skewness in this direction, as shown from other statistical data, is the kind that would arise if the volumes of the cysts measured were normal. But as I have as yet no method for determining their volume, this is only conjecture and not an absolute answer to the query.

The accompanying diagram (text-fig. 2) represents graphically the variation between areas of a single series of 100 cysts measured by two methods: namely, the polar planimeter method and areas based on linear

units, the radius of which is determined from the "size" found by averaging the greatest length and the greatest width. The accuracy of measuring areas by the planimeter method is known to be very great. If the picture of the areas obtained by the linear measurements were equally accurate, the resulting points would cluster about a straight line. If the cluster fails to form this straight line, the deviation is due to the fact that the linear measurements do not show the existing variation in form that the cysts exhibit.

There is, however, a fair degree of correlation between the two methods of measurements. Correlation between two variables is measured by a coefficient

$$*r = \frac{\frac{S(xy) - (m_x)(m_y)}{100}}{S.D._x \times S.D._y}$$

Substituting the values,

$$r = 0.775 \pm 0.03$$

* S = summation; x = the areas as measured by the planimeter method; y = the areas as measured by the linear method; m = the mean; S.D. = the standard deviation. The same meaning is implied for these terms throughout this study.

As this is a positive correlation, it appears that as one variable increases in value the other variable also increases and *vice versa*. From this one can predict the direction which the trend line will take. This is designated by the regression line. The regression lines are the lines plotting the means of the arrays. In this problem it is necessary to plot the regression line for the y variables only, as those are the ones which are being compared with my own measurements. This is calculated by the following formula:

$$y - m_y = r \frac{S.D._y}{\sqrt{S.D._x}} (x - m_x)$$

Substituting values,

$$y - 70.92 = 0.775 \frac{18.49}{\sqrt{18.84}} (x - 75.36)$$

The regression line was plotted and is drawn on the diagram (text-fig. 2).

In conclusion it may be said that if the two methods of measurements were equally satisfactory, the variation about the trend line should be a small portion of the total variation. The variation of areas based on linear measurements for any group of cysts as measured about the trend line is shown by the following formula:

$$S.D._{ay} = S.D._y \sqrt{1 - r^2}$$

Substituting my values,

$$S.D._{ay} = 18.49 \sqrt{1 - (0.775)^2}$$
$$= 11.68 \pm 0.56$$

That is to say, the variation is 11.68 which is, roughly speaking, about two-thirds of the whole biological variation of the cysts measured, namely, 18.49. Therefore the planimeter areas give a more satisfactory measure of form than the combination of linear measurements used by previous authors to determine the "size" of the individual cysts.

Literature Cited

Dobell. C. 1919.—The amoebae living in man: a zoological monograph. 155 pp. London.

Taliaferro, W. H., and Becker, E. R. 1922.—The human intestinal amoeba, *Iodamoeba williamsi,* and its cysts (Iodine cysts). Amer. Journ. Hyg., 2:188-207.

A NOTE ON THE SKIN-REACTION IN TAENIA INFESTATION *

Susan Griffith Ramsdell

The literature with regard to the immunology of parasitism has had to do largely with the complement fixation and precipitation tests and the Casoni reaction in hydatid disease. Characteristic allergic skin reactions, on contact and experimentally, to extracts of the worm, have been reported for ascarids, and for the pin-worm and crab-louse. For the tape-worm, complement fixation in sera of rabbits treated with extracts of the worm has been reported, but Le Bas (1924) could find no evidence of antibody production.

Five cases, one with tape-worm infestation, two with past history, one with an indefinite, and one of hook-worm infection only, presented themselves during the academic year. The unusualness of this condition and the limitations for study have prompted the presentation of this report, although it is incomplete.

A specimen of *T. saginata,* patient P. K., thirty-five grams wet weight, thoroughly washed, was extracted with 70 cc. buffered saline with cresol overnight. This gave a milky emulsion, showing under the microscope fat droplets and amorphous material. The supernatant fluid would not pass through a medium Seitz filter. This was further diluted with the extracting fluid, 1 : 6 and kept in the ice-box. The remaining material, less than 5 grains, was fanned dry, ground, dried, ether extracted and kept in a desiccator. The ether solution was again milky, the ether nonsoluble part brownish. Extraction was repeated and the combined ether extracts when fanned dry gave a brownish-yellow gummy residue with the characteristics tape-worm odor, described as that of old urine or of a rat's nest (acetamide?) This retention of the odor in the ether-soluble fraction was noted by Schwartz for Ascaris (1921). The extract, taken up in saline, gave no skin reaction, although there was more irritation in normal skins than from the alcohol or water soluble fractions. The residue from ether extraction was then extracted repeatedly with 95% alcohol. When evaporated to dryness, there was left a colorless residue with a sweetish, peach-seed-like odor.

The combined ether and alcoholic extracts used as an antigen with the serum of a patient seven weeks after treatment showed no fixation of complement. Perhaps there had been sufficient time for these

* From the Laboratory of University Health Service, University of Texas. The generous cooperation of Dr. George Bethel is acknowledged.

antibodies to have disappeared, as has been noted in echinococcus disease. The residue after ether and alcoholic extractions was taken up in extracting fluid to a malted-milk-like suspension of 1 : 40. This had a wine-like odor. The water-soluble fraction was Seitz-filtered and used for skin testing.

Skin reactions were of two types. When individuals without history (13 tests) were given intracutaneously an amount of the water soluble fraction sufficient to make a bleb about 3 mm. in diameter, there was a slight reddening and spread of the wheal, which disappeared after 5 to 10 minutes. But after 10 to 20 hours, there appeared an area of induration of a centimeter or more with redness and usually with itching. In individuals with a history there was an immediate reaction of the usual sort seen in asthma and hay-fever testing. This began to disappear after about 20 minutes, and the secondary reaction noted in the normals failed to appear. This last finding was consistent, suggests the production of an anti-toxin, and would appear to be of greater diagnostic value than the first allergic reaction. The essential toxicity of extracts of tape has been shown by Messineo and Calamida (1901), who inoculated rabbits, guinea-pigs and dogs with extracts of the dog tape-worm with resulting exhaustion, jerking, partial paralysis, a lowered temperature, and with subsequent marked lesions of the liver. They do not specify, however, that the animals were normal.

A brief summary of the cases follows:

Case 1.—P. K., aged 20 years, student from Texas-Mexican border. History of infestation for several years. Had had no treatment.

11/3. No ova found.

11/20. Eosinophils, 9%.

11/21. Treatment with recovery of 35 grams T. saginata.

12/3. Tested i.c., with whole extract 1:6. No immediate reaction; none in 24 hours. Eosinophils, 7%.

Complement fixation test, negative.

1/13/27. Tested with (1) water-soluble fraction 1:40, i.c. Some spreading of bleb with slight pseudopod formation with itching. (2) With whole extract 1:6. Irritation as in (1) but without pseudopod-formation or itching. Bled. Serum, Seitz-filtered, transferred to normal skin.

1/14. 24-hour reactions: (1) None. (2) Slight induration and redness.

1/16. Transferred serum sites tested with water-soluble fraction. Positive reaction, not marked.

1/17. Transferred serum sites tested with whole extract. Small wheal. Site of 1/16 retested with water-soluble fraction: no reaction.

1/17. Eosinophils, 5%.

1/18. Transfer sites, 24 hour reaction: Retested site: no reaction. Water-soluble site: induration, redness, itching. Whole extract site: as in water-soluble site. Control site: as in water-soluble site.

5/13. Tested with (1) water-soluble fraction 1:20. Distinct and typical reaction in 3 minutes. None in 24 hours. (2) Whole extract, 1:6, as in (1), but giving sl. reaction in 24 hours. (3) Water-soluble, 1:40, as in (1). (4) With powder: negative.

5/28. Eosinophils, 3%.

Case 2.—L. J. W., 22 years old, East Texas. History of tapeworm and hookworm infestation as child. Treated for both at 13 years of age, and hookworm treatment repeated four weeks ago. No ova or segments found.

1/6. Hb., 85% (Newcomer), eosinophils, 6%. No. ova or parasites found in feces.

1/7. No ova or parasites found in feces.

1/26. Eosinophils: 9%. Taenia extract, whole, 1:6. Immediate reaction. 0.1 cc. water-soluble fraction: very much more extensive reaction; hyperemia over whole face of upper arm.

1/27. 24-hour reaction: no induration, only slight area of discoloration. Scratch-test with powdered material: typical pseudopod and redness, about one-fourth that of the l.c. water-soluble-fraction-reaction. Bled for transfer to normal skin.

1/27. No ova or parasites found.

1/29. No ova or parasites found.

1/30. Treated with male-fern, no parasites recovered. (Incomplete collection?)

2/1. Seitz-filtered, slightly tinged serum transferred to normal skin, 2 loci.

2/3. Sites of 2/1 tested with water-soluble fraction 1:10. Reaction four times size of control in untreated area, with severe itching.

2/5. Tested with water-soluble-fraction 1:40. Reaction marked, fading rapidly after 30 minutes. Eosinophils, 6%. Hb, 87% (Newcomer).

Case 3.—T. J. F., Southeast Texas coastal plain. Doubtful history of having passed *Taenia nana* segments three years previously. No treatment.

1/21/27. Eosinophils, 26%.

1/24. Hookworm ova and larvae of *Strongyloides intestinalis.*

1/29. Tested with whole extract and water-soluble fraction: distinctly positive reactions to both, but first without itching or pseudopod formation.

1/30. Thymol treatment: no parasites recovered (incomplete collection). Characteristic tapeworm odor. Few hookworm ova. Hb, 92 (Newcomer).

2/18. Reaction as in 1/29.

3/10. Eosinophils, 18%.

5/9. Rahbditiform larvae *Strong. intest.* found. No ova.

5/14. Reactions as in 1/29, but none to alcohol and ether-soluble fractions. Definite small reaction to scratch test with dry powder.

5/15. 24-hour reaction to water-soluble fraction: slight induration.

5/21. No ova; two Strong. larvae.

5/22. Male fern treatment. No segments recovered.

Case 4.—McM., Austin. History of tape infestation and treatment 6 years previously.

5/28. Tested with water-soluble fraction 1:40. Reaction weak. Eosinophils, 5%.

5/29. 24-hour reaction: small area of induration. No itching.

Case 5.—B. P., Texas-Mexican border. History of treatment for hookworm at 6 years of age in the British Honduras.

5/27. Eosinophils, 20%.

5/28. Definite reaction to *T. saginata,* water-soluble fraction.

5/28. One hookworm ovum found by flotation method.

6/1. Treatment. No parasites recovered. (Very light infestation; hookworm lost?)

A study of these five cases suggests that the antibody response to infestation is of two sorts: (1) an antitoxin production, which diminishes with time following the removal of the parasites from the host; and (2) a sensitization antibody which is apparent after the removal of the parasite. This appears irregularly, as happens in other cases of sensitization; is demonstrable for as much as six years,

and is transferable in the serum to a normal skin. There is evidence of a toxin for normal skin-tissue both in the whole-worm extract and in the water-soluble fraction. The immediate reaction was positive as well in two cases of hookworm infection, one of which also had a Strongyloides infection. No explanation is suggested for this non-specific manifestation.

When the various fractions of the tapeworm were tested for hemolysin and agglutinin against human red cells, it was found that in the whole aqueous extract there was only slight hemolysis after a half hour at 37°C. and twenty hours in the ice-box. In the water-soluble fraction there was no change in the red cells. Both the ether and alcohol soluble fractions, when fanned dry and their residues taken up in saline, gave marked coagulation-like and hemolytic changes. But on using controls of red cells containing 0.05 cc. ether and alcohol respectively, the same changes typical for the solvent were noted. It was therefore not clear that the apparent hemolytic action in the ether and alcohol soluble fractions was due to a true hemolysin since such an action could have been a result of absorbed or loosely combined ether or alcohol.

There is evidence of the occurrence of a skin sensitization and of an anti-toxin production in tape-worm infestation; but the skin reaction is not strictly specific, and the toxins produced are not active as hemolysin or agglutinin.

References Cited

LeBas, G. Z. L. 1924.—Experimental Studies on *Dibothriocephalus latus* in Man. Jour. Helminthol., 2:151.

Leidy, Jos., II. 1924.—The Hematoxins of Intestinal Parasites. A Critical Summary with Notes on Some Cases. Jour. Parasitol. 10:148.

Messineo, E. and Calamida, D. 1901.—Ueber das Gift der Taenien. Centralbl. Bakt., Abt. I, 30:346.

Ransom, B. H. 1922.—Observations on the Toxic Effects of Ascaris Fluids. Jour. Parasitol., 9:42.

Schwartz, B. 1921.—Hemotoxins from Parasitic Worms. Jour. Agric. Res., 22:279.

EARLY DEVELOPMENT OF *DIPHYLLOBOTHRIUM LATUM* IN NORTHERN MINNESOTA*

HIRAM E. ESSEX

For nearly three months during the past summer (June to September, 1927), I had the opportunity of carrying on studies on the development of the broad fish tapeworm (*D. latum*) at Ely, Minnesota. The work was done with the aid of a grant given Dr. Henry B. Ward by the Committee on Medical Research of the American Medical Association. Especial mention must be made of the valuable assistance rendered by Dr. Owen W. Parker and Dr. J. E. Thompson of Ely.

Fecal samples were obtained from individuals who were suspected of harboring the tapeworm. From such samples sufficient eggs were secured to make possible extensive experiments on the early life history. The feces were macerated in tap or lake water, screened, washed and decanted repeatedly until the eggs were as clean as possible. Some eggs were also separated by means of a centrifuge. The egg (Text fig. 1) measured from 70 to 75μ by 46 to 52μ. According to Leuckart (1886) those of the European form measure 50 to 56μ by 35 to 40μ. In other respects the eggs obtained answered the descriptions of those of the European *D. latum*.

At first experiments were planned to determine by what treatment and under what conditions the eggs would hatch best. The eggs obtained by washing and decanting were kept separate from those concentrated by centrifuging. Some cultures were placed in tap water, others in lake water; some were placed in the dark, others in the light. To three cultures 2 per cent formol was added. All cultures hatched equally well irrespective of the treatment or the conditions under which they were incubated, with the exception of those to which formol had been added. None of the eggs so treated gave rise to a single coracidium or ciliated larva. The incubation period ranged from 9 to 12 days but the majority of the cultures required 11 days. Exclusive of the cilia the coracidia (Text fig. 2) measured from 40 to 55μ in diameter. Janicki and Rosen (1917) give the diameter as 42 to 55μ. Nothing was observed in the structure or behavior of the larva which differed from the description given by Janicki and Rosen (1917).

Since *Cyclops strenuus* was identified by Janicki and Rosen as the first intermediate host of *D. latum* in European waters, an attempt was made to find that species of copepod among the plankton forms inhabiting the lakes in the vicinity of Ely. However, that species was not found in

* Contribution from the Zoological Laboratory of the University of Illinois, under the direction of Henry B. Ward, No. 309.

a single instance. Although very common in Europe *C. strenuus* is exceptionally limited in its recorded distribution in the United States. As *C. strenuus* was not found, feeding experiments were performed on all the species of Cyclops obtainable. To fingerbowls containing about a hundred copepods several hundred coracidia were added. Examinations of the Cyclops were made at intervals over a period of 3 weeks.

Among 110 *C. brevispinosus* examined all were negative except 4. Of these 3 contained 1 larva each in the body-cavity. The fourth individual which was examined a few hours after the feeding of the coracidia, had in its intestine two oncospheres from which the ciliated covering had been removed, and one coracidium which was still intact and spinning around within the intestine by means of its cilia. In the cases where the oncospheres had gained the body-cavity, little growth had taken place from 4 to 7 days after the feeding of the coracidia. When removed from the host these larvae measured only 55 to 61μ in length. The same condition was observed among the larvae from the 3 positive *C. prasinus*. The following is a record of the Cyclops which were fed and examined.

Name	No. Examined	Positive	Negative
C. brevispinosus	110	4	106
C. bicuspidatus	132	0	132
C. leuckarti	91	0	91
C. prasinus	24	3	21
C. serrulatus	13	0	13
C. ater	3	0	3
C. fimbriatus	3	0	3
C. albidus	2	0	2
Total	378	7	371

It was evident from these data that the first intermediate host was not to be discovered among the species of Cyclops which occurred in abundance in the lakes near Ely.

The next experiments were made with species of Diaptomus. Janicki and Rosen report that *D. castor* may also act as a first intermediate host of *D. latum*. With great difficulty these copepods were kept alive in the laboratory aquaria. The coracidia were fed to them after which examinations were made at intervals of a few hours to 14 days. Infection was observed in only one species which was identified as *D. oregonensis*. One individual, examined about five hours after the coracidia had been placed in the aquaria, contained within the body-cavity (Text fig. 3) 2 larvae which measured 57 by 48μ. Another individual fed on the 9th of August and examined on the 16th contained one larva (Text fig. 4) which measured 0.11 mm. long and 0.088 mm. wide. Another fed on the 9th of August and examined on the 23rd contained a larva which measured 0.55 mm. long and 0.11 mm. wide. The cercomer or bladder appendage was fully developed and measured 0.11 mm. long and 0.066 mm. wide. It was ovoidal in form in contrast to the spherical form of the European

specimens reported by Janicki and Rosen. These workers observed the hooks adhering always to the cercomer but in one instance I observed 4 of them on the cercomer and 2 on the posterior region of the body. The anterior end possessed a small cup-shaped invagination. Many calcareous corpuscles were distributed throughout the body. The cuticula showed a covering of bristle-like structures which projected posteriorly (Text fig. 5). In all respects, except the shape of the cercomer and the distribution of the hooks, this larva resembled very closely the procercoid of *D. latum* as figured by Janicki and Rosen (1917, fig. 4).

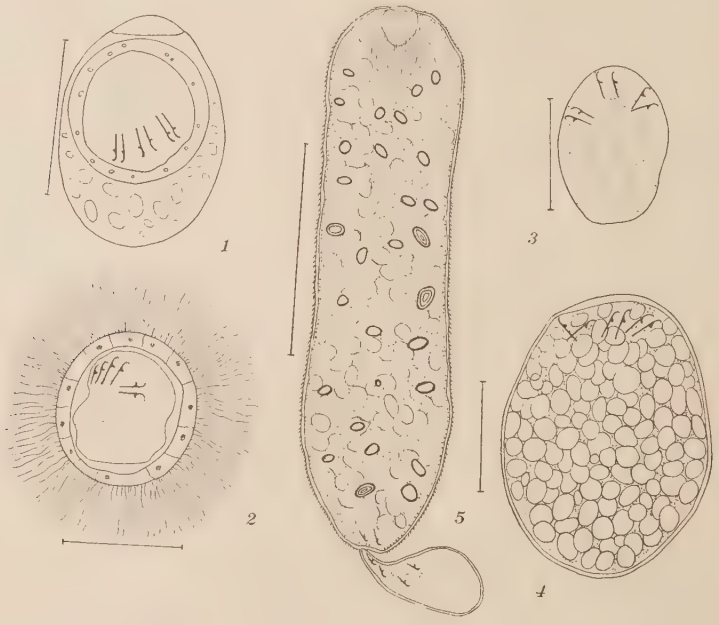


Fig. 1.—Egg incubated 9 days.
Fig. 2.—Coracidium.
Fig. 3.—Larva removed from body-cavity of Diaptomus 5 hours after feeding.
Fig. 4.—Larva after about 7 days' development.
Fig. 5.—Procercoid taken from Diaptomus 14 days after feeding.
Line in Fig. 5 equals 0.2 mm., all others equal 0.05 mm.

A comparison of the studies of Janicki and Rosen with my observations show a very close agreement. They state that the oncosphere when first divested of its ciliated covering measures 24μ in diameter. It measures 24 to 26μ according to my observations. By the 8th to the 12th day they report that the larva measures 0.2 mm. At the end of 6 to 7 days I found the larvae measuring 0.11 to 0.12 mm. in length and about 0.088 mm. wide. Janicki and Rosen state that the larva measures 0 4 mm. an the 14th to the 16th day. By the 14th day I found the length

of the larva to be 0.55 mm. According to Janicki and Rosen no additional structures are differentiated after the formation of the cercomer. I was unable to study a larva which had shed the cercomer but all the structures described by Janicki and Rosen for the mature procecroid were observed in the most advanced larva which I encountered.

References Cited

Janicki, C. and Rosen, F. 1917.—Le Cycle evolutif du *Dibothriocephalus latus*. Bull. Soc. Neuchatel. Sci., Nat., 42:19-53; pls. 1-2.

Leuckart, R. 1886.—Die Parasiten des Menschen und die von ihnen herrührenden Krankheiten. 2 Aufl., 1000 pp. Leipzig and Heidelberg.

A STUDY OF THE REGULARITY OF EGG-PRODUCTION OF *ASCARIS LUMBRICOIDES, NECATOR AMERICANUS* AND *TRICHURIS TRICHIURA**

H. W. Brown

The question whether parasitic worms deposit their eggs at a regular rate is of prime importance in ascertaining the degree of infestation of their host by any egg-counting technique. It is evident at once that if worms produce their eggs irregularly or cyclically the degree of infestation of their host, as diagnosed, will depend to a large extent upon which part of the egg-producing cycle the worms happen to be on the day of examination. Of course in heavy infestations any such cycles of egg laying might be submerged by the overlapping of the individual worm cycles.

Within the last few years it has been shown that several species of parasitic worms pass their eggs at a fairly constant rate. Faust and Khaw (1926) state of the egg-production of *Clonorchis sinensis,* "In a given host, egg-production per worm unit of time is constant"; "Variations in the number of eggs in the stool are due to irregularities in fecal output of the host, to differences in consistency of the stools and to temporary lodgment of eggs in the bile tracts or gallbladder and are not due to actual differences in egg-production per unit time." They also found that recently matured worms produce as many eggs per day as worms that had been mature for a much longer period. Likewise Stoll (1924) found the egg-production of *Necator americanus* to be fairly constant when irregularities due to fecal output were eliminated. The studies recorded in this paper serve to check Stoll's findings in respect to the egg-production by *Necator americanus* and give information on the egg-production of *Ascaris lumbricoides* and *Trichuris trichiura.*

In gathering the data the total daily fecal output from the cases studied was collected. Egg-counts were then made after stirring and weighing. The total number of eggs per day was obtained by multiplying the eggs per gram by the total stool weight. Of the four cases studied, case I was infested with hookworm and Trichuris; case II, with hookworm and Ascaris; case III, with hookworm, Ascaris and

* Contribution from the department of Helminthology of the School of Hygiene and Public Health of the Johns Hopkins University. The work was carried out with the cooperation of the International Health Division of the Rockefeller Foundation. The statistical methods were outlined by Dr. L. J. Reed of the Statistics Department of the School of Hygiene to whom I am greatly indebted for his invaluable suggestions. I wish also to thank Dr. W. W. Cort for help and suggestions.

Trichuris; case IV, with unfertilized Ascaris. Table A presents a summary of the cases studied. The number of female worms harbored by the cases was obtained by dividing the average number of eggs passed per day by the different hosts by the number of eggs a single female of the species has been found to produce, which for Necator is about 9,000 (Stoll 1923a) and for *Ascaris lumbricoides* about 200,000 (Brown and Cort 1927). The relation between the worms harbored and the number of eggs produced has not yet been satisfactorily worked out for Trichuris so no estimate of the number of worms present in the cases studied can be given. The effect the different species of worms may have upon each other's egg-producing capacities when present in the same host is not well understood. The data presented in this paper, however, indicate that the regularity of egg-production of each species is unaffected in multiple worm infestations.

TABLE A—*Summary of Cases Studied*

Case No.	Age	Sex	Infestation	Days Studied	Approximate No. Female Worms	
					Hookworm	Ascaris
I	8	F	Hookworm, Trichuris.....	11	13	0
II	23	M	Hookworm, Ascaris.....	23	135	4
III	8	M	Hookworm, Trichuris, Ascaris....	10	130	20
IV	5	F	Ascaris (unfertilized).....	15	0	2

The weighing and counting were both done by the author thus reducing any errors to a common factor. A dilution of 5 grams of feces to 75 cc. with $\frac{N}{10}$ sodium hydroxide was used in egg counting as described by Stoll (1923, 1924). Three to five 0.075 cc. drops were counted from each specimen.

STATISTICAL METHOD

In treatment of data which includes two variables, that is, in fecal output of the host and any irregularity of egg-production by the worms, certain statistical methods and concepts are essential, due to the necessity of separating these two components and inquiring into the effect of each. If each day's passage of feces represented a single day's accumulation and hence a single day's egg output of the worms, this matter would be very simple. However, with temporary constipations and variations in the completeness of bowel evacuation, the problem takes on a more complicated aspect. Although one day's fecal output does not usually represent the waste accumulation of a single twenty-four hours it is evident that an average of three days' output will more closely approximate three twenty-four hour periods of accumulation within the bowel and consequently three days of egg-production by the worms harbored. It is also evident that the number of eggs produced by an intestinal parasite may vary more or less from day to day

and when the additional variation due to inconstant fecal output is superimposed on this a rather large total variation may be found. The standard deviation (σ) gives a good measure of the variation of the daily egg-production about the mean egg-production over the period of study. By means of three day moving averages of the same egg-output data one would expect to remove some of the variation in egg-output due to inequalities of feces passed and reduce the standard deviation of the daily counts by this simple computation, by the square root of three. This is all the reduction in standard deviation that could be expected if the worms were laying irregularly; any further reduction in the standard deviation by the use of three-day moving averages can therefore only be explained on the basis of regularity of egg-production by the worms.

In obtaining the standard deviation the following formula is used:

$$\sigma = \sqrt{\frac{\Sigma x^2}{n} - M^2}$$

in which M is the mean, n the number of individuals, and x the magnitude of the measurements (eggs per day in this case). Hence for analyzing the data a comparison of the standard deviation of the day to day egg-production with that of the three-day moving averages gives a valuable clue to the rate of egg-production. Four or five day moving averages emphasize this point further by an increased elimination of variation due to fecal output but the three day moving averages proved adequate, except in one case, for a satisfactory comparison of the data presented here.

The second statistical yardstick of value in separating the variations in egg-production due to the variability of the worm from that due to fecal output is their coefficients of variation, which are merely an expression of the standard deviation as a percentage of the mean and are therefore, available in comparing the degree of variation between samples possessing different mean values. It is expressed by the following formula

$$C V = \frac{100\,\sigma}{M}$$

in which σ is the standard deviation and M is the mean. By this means one can compare the deviation about the mean, of the daily egg-production with the deviations about the mean of fecal output and form some judgment on their relative sizes. To establish the validity of the statistical constants obtained from these data their probable errors were computed with the aid of Pearson's Tables for Statisticians and Biometricians. All calculations are made in terms of thousands of eggs which simplifies computation and puts all data on a comparable basis.

Table B presents the data of Case I as secured. The feces for the eleven days studied were all of a formed consistency and were rather constant in amount passed per day. Considering first egg production by Trichuris, I find the mean daily egg output to be, in thousands, 1202. The individual daily counts do not vary widely from their mean and indicate a regular egg production by Trichuris. Three-day moving averages of these same counts give a picture of very regular egg production.

TABLE B—*Original Data of Case I for 11 Consecutive Days*

Day of Study	Stool: Consistency	Stool: Grams	Eggs per Gram: Hookworm	Eggs per Gram: Trichuris	Eggs per Day: Hookworm	Eggs per Day: Trichuris	Eggs per Day, 3-Day Moving Average: Hookworm	Eggs per Day, 3-Day Moving Average: Trichuris
1	Formed*	80	1,200	12,260	96,000	980,800		
2	Formed	108	1,400	14,720	151,200	1,589,760		
3	Formed	89	1,450	16,450	129,050	1,464,050	125,300	1,345,000
4	Formed	89	1,240	10,860	110,360	966,540	130,000	1,340,300
5	Formed	83	1,020	12,640	84,660	1,049,120	108,000	1,160,000
6	Formed	93	1,600	13,200	148,800	1,227,600	114,700	1,081,300
7	Formed	91	1,060	13,800	94,480	1,255,800	110,000	1,177,700
8	Formed	81	1,300	14,450	105,300	1,170,450	116,700	1,218,000
9	Formed	80	1,235	16,660	98,800	1,332,800	100,000	1,253,000
10	Formed	118	1,200	9,270	141,600	1,093,860	115,300	1,193,000
11	Formed	118	1,200	9,270	141,600	1,093,860	127,700	1,173,700
Mean.....		93.6			118,500	1,202,400	116,400	1,216,500

* Stool classification: F, formed; S-F, soft-formed; M, mushy.

TABLE C—*Summary of Data from Case I, Showing Mean Daily Egg Output in Thousands*

	Series Showing 3-Day Moving Average	Mean ± P. E. of Eggs per Day	S. D. ± P. E. of Eggs per Day	Expected S. D.	C. V. ± P. E.: Grams Stool per Day	C. V. ± P. E.: Eggs per Day
Trichuris.....	Day to day, 11 days.....	1202.4 ± 40.2	188.5 ± 28.4	108.8 ± 16.3	15.0 ± 2.3	15.7 ± 2.4
	3-day moving average.....	1216.5 ± 19.1	79.6 ± 13.4			
Hookworm...	Day to day, 11 days.....	118.5 ± 4.9	23.5 ± 3.5	13.6 ± 2.0	15.0 ± 2.3	19.8 ± 3.1
	3-day moving average.....	116.4 ± 2.2	9.4 ± 1.5			

CONSIDERATION OF ORIGINAL DATA

The standard deviation about the mean of the day to day egg-counts is 188.5 ± 28.4 (Table C) while that of the three-day moving average is only 79.6 ± 13.4. However, since the use of the three-day moving average would automatically decrease the standard deviation of the daily output by the $\sqrt{3}$ the comparison of the standard deviation is made between $\frac{188.5 \pm 28.4}{\sqrt{3}} = 108.8 \pm 16.3$ and 79.6 ± 13.4. If the egg-production by Trichuris in this case was unrelated to fecal production by the host no reduction in standard deviation below 108.8 ± 16.3 would have been likely by the use of three-day moving averages. The fact

that a much further reduction in standard deviation was noted indicates that there is a relatively regular egg-production by the worms.

A further proof can be obtained from a comparison of the coefficients of variation of the eggs per day and the grams of feces per day. The coefficient of variation of eggs per day reflects a double variation, due to that of fecal output of the host and any irregularity in egg-production by the worms. Now when these two coefficients of variation are compared it is found (Table C) that of the grams of feces per day to be 15.0 ± 2.3 while that of the eggs per day to be only slightly higher, 15.7 ± 2.4. As this last figure is the C V of the summation of the two variables and we have measured the one, namely fecal output and find it to be practically as large as the combined C V of egg and feces production—the only conclusion possible is that in this particular case there is little variation in egg-production by the worms. Were egg-production as variable as fecal production by the host and the two uncorrelated, the expected coefficient of variation of eggs per day, according to statistical formula, would have been 21.0 ± 2.4.

The hookworm egg-output from this case taken day by day is slightly irregular but the three day moving averages, taking out variations due to fecal passage by the host, give a picture of very regular egg production. The standard deviation (Table C) of the three-day moving averages was 9.4 ± 1.5 while that of the day to day egg output was 23.5 ± 3.5 which when reduced by dividing by the $\sqrt{3}$ gives an expected standard deviation due to three-day averaging of 13.6 ± 2.0. The difference between the actual standard deviation and that which would be expected from random material is significant and indicates regularity in egg-production by the hookworms.

Tables D and E give the data on case II. The egg output in this case was studied over a period of 23 days and indicated a rather heavy infestation with hookworm but light Ascaris infestation. The stools were all of a hard formed consistency and passed with considerable irregularity, several days sometimes elapsing between stools. As this case was one of our technicians it is unlikely that any stools were passed other than those recorded. Due to the extreme irregularity in defecation the egg-output from day to day was exceedingly variable. When three-day moving averages of the eggs per day were made the variation in eggs per day is markedly reduced. Since even three-day moving averages were not adequate to overcome the extreme variation in fecal output it was found necessary to form six-day moving averages to eliminate this factor. This done it is possible to draw some conclusion as to the regularity of egg-production by the worms.

Table E gives the expected and actual reduction in standard deviation due to six-day moving averages. In the case of both hookworm and Ascaris these reductions, which are 430 ± 60.5 to 196.6 ± 19.6 and

271.6 ± 27.0 to 188 ± 18.7 respectively, indicates that once irregularity in fecal output was eliminated, the egg-production by the worms is a fairly regular process. The coefficients of variation of eggs per day (Ascaris, 83.3 ± 12.8 and hookworm, 86.4 ± 13.6) were even lower than that of grams of feces per day (90.0 ± 14.5). This difference is not significant however in light of their probable errors. The fact that

TABLE D—*Original Data of Case II for 23 Consecutive Days*

Day of Study	Stool		Eggs per Gram		Eggs per Day		Eggs per Day, 6-Day Moving Average	
	Consistency	Grams	Hookworm	Ascaris	Hookworm	Ascaris	Hookworm	Ascaris
1	Formed	103	16,340	12,100	1,683,020	1,246,300		
2	Formed	106	15,940	10,640	1,689,640	1,127,840		
3	Formed	119	15,700	10,500	1,868,300	1,249,500		
4	Formed	180	14,920	8,000	2,685,600	1,440,000		
5	Formed	0	0	0	0	0		
6	Formed	0	0	0	0	0	1,321,100	844,000
7	Formed	91	19,320	12,400	1,758,120	1,128,400	1,333,600	824,300
8	Formed	0	0		0	0	1,052,000	636,300
9	Formed	34	23,000	10,700				
		72	22,060	10,720	1,370,320	1,035,640	939,000	606,400
10	Formed	40	17,320	9,600	692,800	384,000	636,800	424,600
11	Formed	185	18,700	11,440	3,459,500	2,116,400	1,214,400	777,300
12	Formed	0	0	0	0	0	1,214,400	777,300
13	Formed	0	0	0	0	0	920,500	589,300
14	Formed	7	18,400	15,900	128,800	111,300	942,000	607,800
15	Formed	119	18,200	12,400	2,165,800	1,475,600	1,074,500	681,100
16	Formed	59	20,900	13,600	1,233,100	802,400	1,164,500	750,800
17	Formed	78	16,260	11,400				
		110	14,660	9,520	2,880,880	1,936,400	1,068,000	720,800
18	Formed	0	0	0	0	0	1,068,000	720,800
19	Formed	64	20,800	12,000	1,331,200	768,000	1,289,800	848,800
20	Formed	76	23,800	18,100	1,808,800	1,375,600	1,569,800	1,059,400
21	Formed	0	0	0	0	0	1,209,000	813,500
22	Formed	54	21,400	19,600	1,155,600	1,058,400	1,196,100	856,100
23	Formed	90	12,720	11,320	1,144,800	1,018,800	906,800	703,300
Mean.....		68.9			1,219,500	798,600	1,119,500	735,500

TABLE E—*Summary of Data from Case II, Showing Mean Daily Egg Output in Thousands*

	Series Showing 6-Day Moving Average	Mean ± P. E. of Eggs per Day	S. D. ± P. E. of Eggs per Day	Expected S. D.	C. V. ± P. E. Grams Stool per Day	C. V. ± P. E. Eggs per Day
Hookworm...	Day to day, 23 days.....	1219.6 ± 148.2	1053.6 ± 104.8	430.0 ± 60.5	90.0 ± 14.5	86.4 ± 13.6
	6-day moving average.....	1201.1 ± 32 2	196.6 ± 19.6			
Ascaris.....	Day to day, 23 days.....	798.7 ± 93.6	665.5 ± 66.2	271.6 ± 27.0	90.0 ± 14.5	83.3 ± 12.8
	6-day moving average.....	768.2 ± 29.1	188.3 ± 18.7			

the coefficients of variation are practically of the same size indicates, however, little irregularity in egg-production by the worms. For, as was emphasized in the discussion of case I, the coefficient of variation of the eggs per day includes the variation due to irregularity of fecal output. When this variation is subtracted from that of the eggs per day variation, there remains no variability due solely to any irregularity of egg-production by the worms.

Tables F and G present the data for Case III. Stools were collected over a period of 10 days. A very large hookworm and Ascaris egg-count was met with in this case while the Trichuris egg count was much smaller. Although on two days no stools were passed by this case the egg output on the day previous and following were greatly increased so that three-day moving averages give a picture of rather regular egg production. This is further emphasized by the great reduction in standard deviation of egg-output of the three-day moving averages

TABLE F *Original Data of Case III for 10 Consecutive Days*

Day of Study	Stool		Eggs per Gram			Eggs per Day		
	Consistency	Grams	Hookworm	Ascaris	Trichuris	Hookworm	Ascaris	Trichuris
1	Mushy	140	8,600	31,520	2,040	1,204,000	4,412,800	285,600
2	Soft-formed	91	14,260	55,230	3,060	1,297,660	5,028,600	278,460
3		0						
4	Formed	86	14,720	66,800	3,320	1,265,920	5,744,800	285,520
5	Formed	108	21,200	75,920	4,260	2,183,600	7,727,060	438,780
6		0						
7	Mushy	129	10,660	31,720	1,320	1,375,140	4,091,880	170,280
8	Formed	61	16,400	46,000	3,000			
	Formed	51.5	21,900	61,500	4,000	2,128,250	5,973,250	391,000
9	Formed	108	9,800	29,600	1,920	1,058,400	3,196,800	207,360
10	Mushy	109	11,100	32,400	1,100	1,209,900	3,531,600	119,900
Mean...	.....	87.8				1,172,200	3,970,800	217,700

TABLE G—*Summary of Data from Case III, Showing Mean Daily Egg Output in Thousands*

	Series Showing 3-Day Moving Average	Mean ± P. E. of Eggs per Day	S. D. ± P. E. of Eggs per Day	Expected S. D.	C. V. ± P. E. Grams Stool per Day	C. V. ± P. E. Eggs per Day
Hookworm...	Day to day, 10 days.....	1172.2 ± 155.2	690.4 ± 107.2	398.6 ± 61.9	52.8 ± 10.5	58.9 ± 12.2
	3-day moving average.....	1165.8 ± 58.7	230.4 ± 41.5			
Ascaris.....	Day to day, 10 days.....	3970.8 ± 526.9	2343.6 ± 372.6	1353.1 ± 215.1	52.8 ± 10.5	59.0 ± 12.2
	3-day moving average.....	3967.2 ± 126.2	494.8 ± 89.2			
Trichuris.....	Day to day, 10 days.....	217.7 ± 31.7	140.9 ± 22.4	81.4 ± 12.9	52.8 ± 10.5	64.8 ± 14.0
	3-day moving average.....	218.1 ± 7.4	27.6 ± 5.0			

(Table G) as compared to the expected reduction in standard deviation due to three-day averaging of random material. Thus the actual standard deviations of the daily output after three-day moving averages were made were $\frac{1}{2}$, $\frac{5}{13}$, $\frac{1}{3}$ the expected S D for hookworm, Ascaris and Trichuris, indicating that once the irregularity of fecal output is eliminated the egg-production is a fairly constant function. The coefficients of variation (Table G) of the eggs per day, which as has been emphasized before is influenced by irregularity of fecal output as well as any irregularity in egg-production by the worms, is not significantly diffeernt

from the coefficient of variation of the grams of feces produced per day. This is further proof of the fact that egg-production by the worms is very regular.

About 25 per cent of the Ascaris eggs passed by this case were of the unfertilized type. This ratio of unfertilized to fertilized egg was very constant, varying from 25 to 30 per cent of each days total egg-production. That these eggs were produced by unfertilized females is indicated by the fact that in no other cases studied were unfertilized eggs found in such large numbers or regularity. Further on treatment of this case females containing only unfertilized eggs were found. These data indicate that unfertilized female Ascarids lay as regularly as the fertilized ones. Case IV which had only unfertilized Ascarids adds further proof of this.

TABLE H—*Original Data of Case IV for 15 Consecutive Days*

Day of Study	Stool Consistency	Grams	Eggs per Gram Ascaris Unfertilized	Eggs per Day Ascaris Unfertilized	Three-Day Moving Averages of Ascaris Eggs per Day
1	Mushy	161	4,500		
	Mushy	30	800	748,500	
2	Formed	188	3,320	624,160	
3	Formed	134	3,800	509,200	627,000
4	Formed	174	2,000	348,000	494,000
5	Soft-formed	167	3,080	514,360	457,000
6	Formed	128	4,460	570,880	478,000
7	.....	0	.....	.....	362,000
8	Formed	131	6,060	793,860	455,000
9	Formed	32	4,640	148,480	314,000
10	Formed	137	5,100	698,700	547,000
11	Formed	81	6,800	550,800	466,000
12	Formed	117	3,600	421,200	557,000
13	.....	0	.....	.....	324,000
14	Mushy	130	6,400	832,000	418,000
15	Mushy	154	2,840	437,360	423,000
Mean.....		117.6		478,800	456,000

In this case as well as the other cases with multiple infestations it will be noted that high egg output from the different species of worms usually occurred on the same day, likewise low egg counts were usually noted from the different species of worms on the same day. Such correlation between the egg output of the different species harbored indicates further that the fluctuation in egg-counts is largely a matter of host irregularity in fecal output. For were the worms laying irregularly or in cycles it would follow by the laws of chance that a high egg-output by one species of worm would frequently come on the same day as a low egg output by another species.

Case IV was studied over a period of 15 days. All Ascaris eggs of this case were of the unfertilized type and this lends it an especial interest, as it sheds some light on the question as to whether unfertilized females function as do fertilized ones in the matter of egg-production. Although on two days no stools were passed (Table H) the egg-production as evidenced by the three-day moving averages is fairly

regular. The standard deviation (Table I) due to three-day averaging is 87.3 ± 11.5 while the expected standard deviation due to this calculation is 147.0 ± 18.1. Likewise the coefficients of variation of eggs and grams feces per day are of the same order, while if the egg production was as variable as the feces output they would be of very different order. The only valid conclusions from these constants is that the egg-production by the unfertilized Ascarids is very regular.

From the foregoing consideration of the data and analysis presented it is seen that any attempt to define or limit the regularity of egg-production by parasitic worms is a matter of some complexity. It involves the elimination of irregularity of egg-output due to variations in the host's fecal output from day to day. In defining regularity of egg production by worms a standard of comparison is of course necessary. The statement that worms produce eggs at a fairly constant rate does not give a particularly clear idea of just what is meant. Since

TABLE I.—*Summary of Data, Case IV, Showing Mean Daily Egg Output in Thousands*

	Series Showing 3-Day Moving Average	Mean ± P. E. of Eggs per Day	S. D. ± P. E. of Eggs per Day	Expected S. D.	C. V. ± P. E. Grams Stool per Day	C. V. ± P. E. Eggs per Day
Ascaris..... (unfertilized)	Day to day, 15 days.....	479.8 ± 44.3	354.6 ± 31.4			
	3-day moving average.....	455.5 ± 14.1	87.3 ± 11.5	147.0 ± 18.1	53.1 ± 8.2	51.4 ± 7.8

egg-output is so closely linked with fecal output of the host, a comparison of these two seems to afford a good standard for judging the regularity of egg production. To be able to say that in general the egg-production by parasitic worms is a fairly constant phenomenon and to limit it still further by stating that it is far more regular than fecal output by the host is about as close as this study shows the egg production can be gauged. When the fecal output of the host is regular it is evident from the eggs passed per day that the worms produce them at a very regular rate. This was emphasized in Case I of this study in which daily variations in egg-production were very small.

In general it appears that all three species studied produce eggs with about equal regularity. Differences in regularity of egg production by the species in the cases studied can be in part ascribed to size of infestations. Thus with a light infestation variations due to missing eggs when egg-counting affects the final result much more than in a heavy infestation. It is interesting to note that the large *Ascaris lumbricoides* laying in the neighborhood of 200,000 eggs per day (Brown and Cort, 1927) apparently produces eggs as regularly as *Necator americanus* which produces about 9,000 eggs per day. This study

indicates that egg-counts of Ascaris and Trichuris are a valid measure of the degree of infestation of the host and that any variation in day to day egg-output is a matter of irregularity of the hosts functioning rather than irregularity of worm functioning. Stoll's finding in regard to regularity of egg production by Necator are confirmed with the same conclusion.

SUMMARY

1. Studies of the egg-production are presented from four individuals of *N. americanus, A. lumbricoides* and *T. trichiura,* covering a period from 10 to 23 days.

2. Judged by a comparison of the coefficients of variation of the eggs per day and grams feces passed by the host, egg-production is a very constant phenomenon, much more so than feces passage by the host.

3. From a comparison of the standard deviations of day to day egg-outputs with those of three-day moving averages of the same data once irregularities due to fecal production are eliminated egg-production is fairly regular.

4. Egg production by *N. americanus, A. lumbricoides* (fertilized and unfertilized) and *T. trichiura* is, within errors due to size of infestations, equally regular.

5. Egg-count data from hosts harboring these worms give an index of the number of worms harbored.

6. Surveys of population groups by egg-count give as reliable information for *Ascaris lumbricoides* and *Trichuris trichura* as for *Necator americanus.*

REFERENCES CITED

Brown, H. W. and Cort, W. W. 1927.—The egg production of *Ascaris lumbricoides.* Jour. Parasit., 14:1-15.

Faust, E. C. and Khaw, O. 1926.—The egg-laying capacity of *Clonorchis sinensis.* Proc. Soc. Exp. Biol. and Med., 22:606-607.

Stoll, N. R. 1923.—Investigations on the control of hookworm disease. XV. An effective method of counting hookworm eggs in feces. Amer. Jour. Hyg., 3:59-70.

1923a.—Investigations on the control of hookworm disease. XVIII. On the relation between the number of eggs found in human feces and the number of hookworms in the host. Amer. Jour. Hyg., 3:156-179.

1924.—Investigations on the control of hookworm disease. XXIII. The significance of egg count data in *Necator americanus* infestations. Amer. Jour. Hyg., 4:466-500.

OBITUARY

It is with deep sorrow that THE JOURNAL records the death, September 22, of Sir Arthur Everett Shipley at the Master's Lodge, Christ's College, Cambridge, England. An indefatigable worker in various fields, he contributed significantly to the knowledge of animal parasites and it is hoped to include in an early number a fitting tribute to his work in this field. A man of genial and sympathetic nature, Shipley made a lasting and inspiring impression on all who were fortunate enough to come in personal contact with him. By his death science has suffered the loss of an able worker and those who knew him have been bereft of a true friend.

SOCIETY PROCEEDINGS

ABSTRACTS OF PAPERS CONTRIBUTED FOR THE THIRD ANNUAL MEETING OF THE AMERICAN SOCIETY OF PARASITOLOGISTS, DECEMBER 27-30, 1927, NASHVILLE, TENN.

Some Studies on the Biology of Ancylostoma braziliense *and Its Relation to Creeping Eruption.* W. E. Dove, School of Hygiene and Public Health, Johns Hopkins University.

The developmental period of *Ancylostoma braziliense* in cats and dogs varied from 13 to 27 days. The percentage of worms which developed as a result of skin infections was low in comparison with the infections by mouth. Worms obtained from the dog were carried through four successive generations in dogs, and from each of these generations larvae were cultured which produced lesions of creeping eruption. Worms obtained from the cat were carried through seven generations in cats. In each case where larvae from these animals were tested on man, typical lesions were produced. Larvae from dogs infected with a cat strain, and larvae from cats infected with a dog strain, also produced creeping eruption lesions. When larvae were concentrated on cats and dogs, a "ground itch" was produced which was comparable to that produced on the human skin by larvae of the human hookworms. When the larvae were distributed on the skin of cats and dogs, no lesions or papules resulted. Attempts to produce creeping eruption on laboratory animals have been unsuccessful. Attempts to secure a hybrid with *Ancylostoma braziliense* and *A. caninum* were unsuccessful. Tests made for parthenogenesis gave negative results. The identity of *Ancylostoma braziliense* recovered in Florida and the species encountered in Texas is indicated by interbreeding experiments. From cats harboring each of these strains the immature worms were separated according to sex, and were fed to kittens known to be free of hookworms. Each kitten received the males of one strain and the females of another strain. In due time, infective hookworm larvae were cultured from the feces of each of these kittens. At Dallas, Texas, where the rainfall is less than that of Florida, low incidence of creeping eruption accompanies a light infestation of *A. braziliense* in dogs. Here creeping eruption infections occur on localized spots where the soil is wet. Under such conditions patients are more often infected with multiple lesions.

Further Observations on Creeping Eruption. J. Lee Kirby-Smith, Jacksonville, Fla.; W. E. Dove, and G. F. White, Bureau of Entomology, U. S. Department of Agriculture.

During 10 days of July, 1924 and also of July, 1925, clinics were conducted in Jacksonville, Fla., for the study of creeping eruption. During these more than 300 creeping eruption patients were examined and treated. The observations made were in connection with the symptoms, causation, epidemiology, diagnosis and treatment of the disease. To the layman creeping eruption is best known as being "ground itch." Many of those coming to the clinic were suffering from itchy skin diseases other than creeping eruption. About three dozen of these different diseases were diagnosed. Fungous infections were most commonly encountered. Adults representing 17 different occupations were among the creeping eruption patients. The highest percentage of infections occurred among children between 5 and 7 years of age. Infections were most common on the feet, although it was seen on all parts of the body excepting the scalp. In most instances a history was obtained that the part infected had been in con-

tact with damp soil. Any blistering agent is likely to be effective in treatment, the reasons for this often being possibly that the epithelial layer in case of a blister is split at the level at which the parasite usually occurs. The senior author in his practice has found that treatments with ultra-violet ray that cause an exfoliation of the epidermis has often resulted in prompt cures of extensive cases.

Experimental Studies on Creeping Eruption. G. F. White, and W. E. Dove, Bureau of Entomology, U. S. Department of Agriculture.

Using infective larvae from pure cultures of *Ancylostoma braziliense* obtained from the dog and the cat, the advancing, tortuous, linear lesions that characterize creeping eruption have been produced in the human skin. In making inoculations during the summer it is only necessary to apply the desired number of larvae to the perspiring skin in a small drop of water. A bit of sand or charcoal is added to the drop to hold it in place and possibly to facilitate penetration. A slight pricking sensation may be experienced within 10 minutes; this has been delayed more than two hours in some instances, and, indeed, not observed at all in others. During cool weather heating the skin in connection with moisture seems to facilitate penetration. Invasion by 100 per cent of the larvae used is not to be expected. Usually the first sign to be observed from inspection is a diffuse erythema within 24 hours about the point of ingress of the larva. This is preceded, accompaned, and followed by itching. Soon a papule-like elevation is seen followed, as a rule, within a few days by advancing, linear lesions. In the inoculation of dogs and cats, using larvae, feeding has been the method of choice. When adult worms were used they were usually pipetted into the mouth. Adding the adult worms to a small amount of milk or to fresh ground meat and giving these to hungry animals sufficed well in making the inoculations. The identity of *A. braziliense* obtained at Jacksonville, Fla., from the dog and from the cat was shown by cross inoculations, the experiments being done with both larvae and adult worms.

Notes on the Biology of Trichinella spiralis. Reed O. Christenson, University of Minnesota.

An accidental infection of twenty-seven rats with heavily trichinized rabbit tissue has given additional data relative to the sex ratio of intestinal trichinae. These data run from fifty-five hours, the time the first rat died, well into the sixth week, when the rats showing the symptoms most clearly were killed and examined. The dosage was lethal to most of the rats. Notes are included on a series of experiments to test the possibility of prenatal infection with *Trichinella spiralis,* and on the incidence of this parasite in the Twin Cities as representative of its prevalence in the north-central United States. This work supplements the note published by the author in Science of September 16, 1927.

Some Effects of Cysticercus fasciolaris *Rud. on the White Rat.* Harry M. Miller, Jr., and Charlotte W. Dawley, Washington University, St. Louis.

A study was made to determine in what ways *Cysticercus fasciolaris,* normally parasitic in the liver of wild rats and mice, may affect the white rat. Onchospheres obtained from gravid proglottids of *Taenia teniaeformis* were fed in varying doses. In some cases thousands of cysts developed in the liver; in such instances general effects were observable within ten days. The effects on the blood were especially studied. The erythrocyte count was lower in infected than in control rats; the leucocyte counts were not consistent. The number of eosinophils increased from time of infestation, reaching a high level after fifteen days, and remaining high for ten days in one series of experiments (light infestations) and much longer in another series (heavy infestations). No local eosinophilia was observed in the liver. That the cyst fluid of *Cysticercus fasciolaris* is not very toxic was shown in several ways. Neither repeated intra-

peritoneal injection of cyst fluid nor release of cyst fluid into the abdominal cavity by puncture of considerable numbers of cysts was followed by any effects like those resulting from rupture of Echinococcus cysts in man. Instillation of cyst fluid into the eyes of infested and control rats did not produce a local eosinophilia; nor was an intra-dermal reaction gotten with the use of powdered *C. fasciolaris*. The conclusion is that this cestode is a benign parasite in the white rat.

Platyhelminthes in Insects. E. W. Stafford, Mississippi Agricultural and Mechanical College.

A number of species of parasitic worms are reported from American amphibious insects. Among these are found forms belonging to the genera Hymenolepis, Plagioporus, Eumegacetes, Pleurogenes, Lecithodendrium and others for which the insects were serving as transfer hosts.

Two Types of Oocysts of Coccidia from Iowa Calves. Elery R. Becker, Iowa State College.

Microscopical examinations for protozoa were made of the feces of thirty-four calves. These calves were from one to four months of age, and so far as the writer knows were born either in or near Ames, Iowa, and had never been out of the state. Three of these calves were found to be infected with coccidia of the genus Eimeria, but none of the three suffered any clinical symptoms of dysentery. The coccidia from two of the calves were of the ovoid type with a slightly salmon tint; those from the other calf were ellipsoidal and colorless. The ovoid forms from one calf had a mean length of 29.7 μ, a mean breadth of 20.2 μ. The ovoid forms from another calf were very similar in size. The ellipsoidal forms had a mean length of 23.4 μ and a mean breadth of 15.9 μ. The ratio of mean length to mean breadth in both cases was almost exactly the same; i.e., 1.47. When cultured in one percent potassium dichromate solution both types developed four sporoblasts, in each of which appeared two sporozoites. There was no residual element (Restkörper) in the cyst of either form, but in the spores of both there was a residuum (Restkörper) in addition to the two sporozoites.

Some Notes on Leucocytozoa with special reference to Leucocytozoon anatis. Ernest Hartman, University of Illinois.

L. anatis Wickware 1915 was found both in wild and tame ducks in Emmet and Cheboygan counties, Michigan, in the summers of 1926 and 1927. Tame geese in the same region apparently had no Leucocytozoa which makes it seem likely that *L. anseris* Knuth and Magdeberg 1922 is a distinct species. Two young Coopers hawks *(Accipiter cooperii)* were found to harbor Leucocytozoa which differed in morphology from those in the duck. Cages covered with wire cloth having 18 mesh per inch protected young ducks against infection. Unprotected ducks developed clearly recognizable forms of the parasite in about ten days. The earliest gametocyte stages approach a sphere in form and do not possess the well defined spindle shaped host cell which is characteristic later in the infection. Large dividing forms which were probably schizogenous stages were frequent. About sixty merozoites were found from one schizont on a smear made from the peripheral blood. Contrary to the described characters of the genus Leucocytozoon these parasites definitely contain pigment. From this and from the appearance of the earliest recognizable forms of the parasite it is concluded that the parasite enters young red cells. Experimental infections terminated fatally in some cases. The spleen, liver and kidneys were abnormally large and congested. One case fifteen months after the primary infection showed a marked atrophy of the spleen. Tissue sections gave evidence of degeneration throughout most of the organs of the body.

Studies on the Trematode Family Strigeidae (Holostomidae). Alaria nasuae *sp. nov.* George R. LaRue and Elsie W. Townsend, University of Michigan.

This study is based on four worms found in the intestinal ceca of *Nasua nasica,* comprising lot no. 26151, Bureau of Animal Industry, Washington. A study of sections and *in toto* preparations and comparisons with descriptions and preparations of all known species of Alaria shows this to be a new species which we have named *Alaria nasuae,* the specific name indicating the host. Average length 3.87 mm., average width of forebody 2.44 mm., average width of hind-body 1.081 mm. Hold-fast larger than in *Alaria alata.* Lateral sucking cups present. Oral sucker 0.056 by 0.06 mm., prepharynx lacking, intestinal ceca extending to posterior end of body. Acetabulum 0.052 by 0.048 mm. Testes much lobed, ovary globose, jutting up into fore-body, Laurer's canal present, opening to exterior, ootype near anterior level of T_1, vitellaria confined to forebody and hold-fast organ. Vasa efferentia join vas deferens anterior to T_1, vas deferens inflated, vesicula seminalis a complex mass of coils, ductus ejaculatorius large, muscular. Uterus with extensive transverse loop between T_1 and T_2, storage coils of uterus in hold-fast organ.

Studies on the Trematode Family Strigeidae (Holostomidae). Alaria oregonensis, *n. sp.* George R. LaRue and George H. Barone, University of Michigan.

Material: Lot 26323, Bureau of Animal Industry, Washington, from small intestine (anterior end) of coyote, Redmond, Oregon, collected by E. B. Cram, 15-1-25. The worms were grouped in a long narrow mass at the anterior end of the intestine along the mesenteric side. Adult worms are large and thick-bodied, measuring 6.33 by 8.25 mm. long and 1.45 by 3.4 mm. broad. Constriction between fore- and hind-body regions shallow, bodies rarely flexed. Foliate margins of fore-body tightly rolled about hold-fast and host tissue clamped between these organs. Hold-fast organ long, slender, grooved ventrally, projecting well beyond oral sucker, attached to fore-body by slender peduncle. Oral and ventral suckers weak, acetabullum 0.077 to 0.118 by 0.062 to 0.99 mm., oral sucker 0.074 to 0.095 by 0.059 to 0.035 mm. Lateral sucking cups or tentacular appendages present. Pharynx large, muscular; eosphagus short; digestive ceca long, extending to level of ductus ejaculatorius. Sexual organs as in Alariinae. Ootype lateral to T_1, uterus with large transverse loop between T_1 and T_2, vesicula seminalis large, composed of many coils, ductus ejaculatorius very muscular. Ovary conical, composed of 10 to 12 lobes, oviduct ampulliform, serving as receptaculum seminis. This species appears to be new and is given the name *Alaria oregonensis.*

Studies on the Trematode Family Strigeidae (Holostomidae). Neodiplostomum lucidum *n. sp.* George R. LaRue and Nelly J. Bosma, University of Michigan.

Material: seven worms from the intestine of the opossum, *Didephys virginiana,* collected by Dr. E. W. Price in Texas. Frequence of occurrence, unknown. Preserved material characterized by an unusual transparency, which suggests the specific name *lucidum.* Length of preserved type specimen 1.944 mm., breadth of hind-body 0.333 mm., fore-body leaflike, varying greatly in width and in degree of rolling of the leaflike margins in the several specimens. Lateral sucking cups or tentacular appendages absent. Cuticula of fore-body spinous. Oral sucker 0.111 mm. in diameter, pharynx muscular, esophagus short, intestinal crura descending laterally and ventrally to the posterior end of the body. Acetabulum 0.094 mm. in diameter. Hold-fast organ protruded, elongated oval in shape, longitudinally cleft, and broadly attached to fore-body. Ovary reniform, anterior and dextral to testes. Laurer's canal opening dorsally at level of anterior testis. Vitellaria most abundant in the fore-body in region of the hold-fast organ, follicles less numerous in hind-body. Vitelline reservoir

between testes. Mehlis gland and ootype to left of anterior testis. Uterus ascending barely into fore-body, not penetrating into hold-fast organ. Anterior testis smaller than posterior, and not horseshoe-shaped. Posterior testis horseshoe-shaped. Seminal reservoir anterior to T_1, into which vas efferens from each testis discharges. Seminal vesicle large, consisting of a simple coil; ductus ejaculatorius weakly muscular, opening into common genital passage. Eggs large, few in number. This species is assigned for the present to the genus Neodiplostomum under the name of *Neodiplostomum lucidum* n. sp.

Notes on the Trematode Genus Cryptocotyle. H. W. Stunkard, New York University.

Metacercariae of Crytocotyle occur encysted in the fins and gills of *Tautogolabrus adspersus* at Woods Hole, Mass. On morphological grounds they were described by Linton (1915) as larval stages of *Cryptocotyle lingua* (Creplin), parasitic in the intestine of the loon, gulls, and terns. These larvae were experimentally developed in the white rat and in the cat. Excystment occurred in the intestine of the domestic duck but the worms did not develop full sexual maturity there. Specimens from marine birds did not develop sexually to the extent obtained in experimental mammalian hosts. Frequently living trematodes, sexually immature, were recovered with the droppings of domestic ducks and marine birds. No such specimens were recovered from rats or cats. Dissection of the intestinal wall of ducks, cats, and rats at all stages after feeding disclosed worms between the villi and sometimes partially enclosed in the lumen of the glands, but no penetration into the wall of the intestine as reported by Yokogawa, Ciurea, and Faust and Nishigori for other genera of the family Heterophyidae was observed. The normal flame cell formula for the metacercariae of Cryptocotyle is $2\times(3+7+7)+(7+7+7)$. Some variation in the number and position of the solenocytes was observed.

Methods of Diagnosing Human Endamoeba. Thomas B. Magath and Charlotte B. Ward, Mayo Clinic.

A comparison is made of the diagnosis of Endamoeba by the fresh stool method, the permanent staining method and the culture method with a view to determining the most practical method available for use in a clinical laboratory.

Experimental Amoebiasis in Kittens. Charles W. Rees, School of Hygiene and Public Health, Johns Hopkins University.

These experiments were conducted to secure accurate data on the pathological reactions of the tissues of kittens to the attack of *E. histolytica*. The material consisted of a strain of *E. histolytica* and kittens. The parasites were maintained in tubes of L. E. S. medium to which rice starch was added. The kittens ranged in weight from 400 to 800 grams. To infect them a laparotomy was performed, the colon ligated near the rectum and the culture of *E. histolytica* injected cephalad of the ligature. The kittens were killed and autopsied at intervals ranging from 24 hours to 144 hours after the operation. When infected the colon was examined macroscopically and then fixed and sectioned for microscopic study. Growth of *E. histolytica* was greatly accelerated by rice starch. The rice-fed strain established itself in 7 out of 13 kittens. In one kitten killed at 90 hours the lining epithelium of the colon was completely destroyed, the mucosa and submucosa greatly thickened and many of the cells were necrotic. Numerous endamoebae were found throughout both the mucosa and the submucosa. In another case at 144 hours there was an equally general penetration of the endamoebae but except for this the mucosa and submucosa appeared almost normal. In another case at 48 hours where the contents of the colon were teeming with *E. histolytica* there were no lesions either macroscopic or microscopic. Much of this variability of reaction of the host to the parasite was later

reduced by a uniform method of treatment. The kittens were starved 24 hours and given cathartic pills 6 to 8 hours previously to the operation and the dose of *E. histolytica* standardized. In severe infections it appeared that bacteria played an important role in producing the lesions.

Complement Fixation in the Diagnosis of Infections with Endamoeba histolytica. Charles F. Craig, Medical Corps, U. S. Army, Washington, D.C.

During experiments upon the presence in alcoholic extracts of cultures of *Endamoeba histolytica* of hemolytic and cytolytic substances, it was discovered that such extracts also produced complement-fixation when used as antigens with the blood serum of individuals infected with this parasite. The present paper gives the results obtained in the diagnosis of infections with *Endamoeba histolytica* using such extracts as antigens and the method of making the extracts and of applying the test. The conclusions reached as the result of the examination of over 300 blood sera from as many individuals with the complement fixation test as outlined in the paper are as follows:

1. Complement fixing substances can be demonstrated in the blood serum of individuals infected with *Endamoeba histolytica* by using as antigens alcoholic extracts of cultures of this parasite.

2. The complement fixation reaction disappears in cases of infection with this parasite that have been treated and the endamoebae eliminated.

3. Individuals free from infection with *Endamoeba histolytica* do not give a positive complement fixation reaction when their blood serum is tested with such extracts of cultures of *Endamoeba histolytica.*

4. The reaction does not occur in individuals infested with *Endamoeba coli, Endamoeba nana, Iodamoeba williamsi, Trichomonas hominis* and *Chilomastix mesnili.* It is probable, therefore, that a positive reaction does not occur in other protozoan infections or infestations when such an antigen is employed in the test.

5. The reaction does not occur in individuals suffering from other diseases or infections.

6. The reaction does not occur in individuals giving a positive Wassermann or Kahn reaction if *Endamoeba histolytica* is not present.

7. The reaction occurs not only in individuals presenting definite symptoms of infection with *Endamoeba histolytica* but also in cases presenting indefinite symptoms and in "carriers" without any symptoms of the infection.

8. The practical value of the test in the diagnosis of infection with *Endamoeba histolytica* is still uncertain, owing to the difficulty of preparing suitable extracts, the technical difficulties of the test, and the fact that in practically every case a proper examination of the feces, microscopically and culturally, results in the demonstration of the parasite in practically every case giving a positive complement fixation reaction.

Changes in the Blood of Cats due to Amebiasis. Elizabeth P. Sanders, School of Hygiene and Public Health, Johns Hopkins University.

Kittens were inoculated intrarectally with cultures of *Endamoeba histolytica.* Upon the inoculated kittens, and control animals (uninoculated ones of the same litter) total red and white blood cell counts and differential blood counts were made daily or bi-daily for a week prior to inoculation and during the period of infection. Careful records were kept at the same time of the intestinal parasites found. Results indicated that kittens inoculated with *E. histolytica* will develop acute amebiasis, causing slight anemia, loss iln weight, a marked polymorphonuclear leucocytosis coupled with a rise in the number of large mononuclear cells, terminating in the death of the animal. The polymorphonuclear leucocytosis may be due to secondary bacterial infection.

Changes in the Blood of Cats due to Trypanosomiasis. Justin Andrews and Elizabeth P. Sanders, School of Hygiene and Public Health, Johns Hopkins University.

Litters of young cats were separated into two groups. One group was inoculated with pathogenic trypanosomes *(T. equiperdum* and *T. brucei);* the others were kept for control. Trypanosomes appeared in the blood within two days; the infection, accompanied by the usual symptoms, terminated in death within one and one-half months. During the period of infection and for five days prior to inoculation total red and white blood cell counts, differential blood counts, hemoglobin, and blood sugar determinations were made either daily or bi-daily. Results show a progressive secondary anemia, a pronounced general leukapenia, which seems to affect all the white blood cells in about the same degree, and little change in the blood glucose until just before death when there is an abrupt decline.

Contributions to the Life History of Protecephalus ambloplitis *(Leidy).* George W. Hunter, III, Rensselaer Polytechnic Institute.

During the summer of 1927 an experimental study of the life history of *Proteocephalus ambloplitis* was undertaken for the U. S. Bureau of Fisheries. Adults of *P. ambloplitis* were secured from the upper intestine of the large mouth black bass *(Micropterus salmoides)* and the eggs were placed in aquaria containing Entomostraca. Periodic examinations were made and larvae were recovered from the 5th to 16th day of infection from the body cavity of *Cyclops albidus* and *C. prasinus.* The smallest recovered were about 0.04 mm. in length and the largest taken from the Cyclops on the 16th days of infection was slightly over 0.11 mm. long. Three days after starting the experiment 62 fry *(M. salmoides)* were added to the aquaria. Procercoid larvae were recovered from the stomach wall, mesentery, liver and ovary of the fry. Ten were examined and 50 per cent were infected. Ten days after the introduction of the fry those which were still unexamined were transferred to a larger aquarium and 8 yearling bass *(M. salmoides)* from uninfected stock were added. At the end of 12 hours all the fry had disappeared. Within 3 days four yearlings were examined; three of these yielded young pleurocercoid larvae, about 0.3 to 0.4 mm. in length, possessing evaginated scolices. Control specimens of Cyclops, fry and yearlings were examined.

Life History Studies on Trematodes from Missouri. Oliver R. McCoy, Washington University.

In the course of a two year study of the larval trematodes in the vicinity of St. Louis, Missouri, clues were discovered which led to the establishment of two new trematode life histories, the determination of the metacercarial stages of a furcocercous cercaria, and the identification of four xiphidiocercariae as the larvae of members of the sub-family Reniferinae Pratt. *Echinoparyphium flexum* (Linton 1892) has been developed experimentally in laboratory bred chicks by feeding cysts from *Planorbis trivolvis;* the cercaria has been naturally observed and experimentally obtained in *Physa integra.* A xiphidiocercaria from *Planorbis trivolvis* was found to encyst in crayfish and damsel fly larvae and develop experimentally in the intestine of the catfish, *Ameiurus natalis,* into a new species of *Plagiorchis;* the adult worm also naturally occurs in this host. *Cercaria hamata* Miller 1923 a furcocercous "monostome" larva, was observed to penetrate small sunfish, *(Eupomotis gibbosus),* and develop in the muscles into a larval holostome. Ova from *Renifer kansensis* Crow 1913, *Dasymetra conferta* Nicoll 1911 and *Pneumatophilus variabilis* (Leidy) Odhner 1910, collected from snakes from the vicinity of St. Louis, were used to infest laboratory bred snails, (Physa integra), and the cercariae of these three forms have been obtained. In addition a new cercaria from *Physa integra* was found to encyst in tadpoles and develop

in snakes into young worms which most probably belong to the genus Renifer. These four stylet cercariae are alike in general characteristics, the most striking of which is the excretory vesicle with lateral arms extending anterior to the ventral sucker. They are closely related to *Cercaria brevicaeca* Cort.

A New Bothriocephalid from Diemictylus viridescens *with Notes on the Life History.* Lyell J. Thomas, University of Illinois.

A new species of bothriocephalid tapeworm from the little green newt, *Diemictylus viridescens,* is described. A preliminary study of the life history carried on at Douglas Lake, Michigan, the summer of 1927, indicates that two crustacea, *Cyclops albidus* and *Cyclops brevispinosus* act as intermediate hosts.

Ticks Found on Nigerian Rodents and Insectivores. A. S. Pearse, Duke University.

A statistical study has been made of the ticks found on about 500 small mammals belonging to 30 species. The relations of infestation to habits and habitats of the hosts are discussed.

Viability of Trichomonas hominis *at Various Temperatures and in Various Dilutions of Water.* Robert Hegner, School of Hygiene and Public Health, Johns Hopkins University.

There is no cyst stage in the life cycle of *T. hominis,* hence the trophozoites must be sufficiently resistant to withstand conditions outside the body of the host until entrance is gained into a new host. Heavily infected fecal material was kept at room temperature, summer temperatures, low temperatures and high temperatures. Most of the trichomonads succumbed at room temperature within 48 hours, but a few were still alive at the end of 8 days; these were capable of growth and multiplication in culture media and hence presumably infective to susceptible hosts. Similar results were obtained when summer temperatures and low temperatures were used. All trichomonads died within 5 hours at high temperatures (40°C and 44°C). Infected fecal material was diluted with tap water. The trichomonads were not injured at low dilutions, but most of them died in a short time when dilutions of 5 grams of feces to 95 cc. of water, and 1 gram to 99 cc. were made. No trichomonads lived more than 6 hours in the latter dilution. The results when distilled water and spring water were used as diluents were similar. The death of the trichomonads was apparently due to changes in the osmotic pressure; for example, the osmotic pressure of undiluted feces, obtained by the freezing-point depression method was 6.145, that of the serum-saline-citrate culture medium, 7.469, that of 5 grams of feces to 95 cc. water, 0.483 and of 1 gram to 99 cc., 0.121. It is concluded that trichomonads are able to withstand a wide range of temperatures while outside of the host's body and have a good chance of reaching a new host in a viable condition, but that there is not much danger of infection being due to contamination of the water supply.

The Ingestion of Red Blood Corpuscles by Trichomonad flagellates. Robert Hegner, School of Hygiene and Public Health, Johns Hopkins University.

The presence of red blood cells in trichomonads and other intestinal protozoa has been accepted by various investigators as evidence of pathogenicity; whereas others hold that red blood cells simply serve as an article of food in the same sense that bacteria or organic debris do. No test, however, has been made to determine accurately how frequently red blood cells are ingested by various species of trichomonads. Trichomonads were obtained in serum-saline-citrate cultures from the mouth of man, from the vagina of the monkey, *Macacus rhesus,* from the intestine of the same species, from the intestine of the chicken and from the intestine of the rat. Four drops of medium positive for each

trichomonas were added to each of 21 fresh tubes; then 3 drops of blood from man were added to 3 tubes, 3 drops from the dog to 3 tubes, and similar amounts from the cat, rabbit, rat, guinea-pig, and mouse to 3 tubes each. After incubating at 36°C. for 24 hours material from the bottom of the tubes was examined and records of the first 100 trichomonads found were made with reference to the presence or absence of red blood cells and their number if present. Each species of trichomonad ingested red cells of every species of mammal used. Most of the positive specimens contained one red cell, many ingested 2, and a few contained 3, 4, or 5. One hundred red cells from each mammal used were measured. The mammals used in order of the average preference of their red cells by trichomonads are as follows, the average diameter of the red cells being included in parentheses: rabbit (6.0 μ), mouse (5.4 μ), rat (6.1 μ), cat (5.7 μ), guinea-pig (7.4 μ), dog (7.4 μ), man (7.8 μ). It appears that the smaller red cells were more often ingested than the larger, thus indicating that it may be the size of the red cells and not some other factor that accounts for the differences observed.

Infection Experiments with Amoeba hydroxena *Entz.* Bruce D. Reynolds, University of Virginia.

Amoeba hydroxena is an ectozoic parasite on the fresh-water polyp, Hydra. Upon sectioning infested hydras one frequently finds amoebae in the enteric cavity. This paper deals with two phases of experimental infection: I. (*a*) Pathogenicity of the amoebae when the host is well fed, and when the host is starved. (b) Resistance of the parasite to the enteric juices. II. The ability of the amoebae to attack other forms, notably Stenostoma and Microstoma.

The Separation of a Tritrichomonas of Man from Bacteria and Its Failure to Grow in Media Free of Living Bacteria and in the Blood Stream of Warm- and Cold-blooded Vertebrates. L. R. Cleveland, Harvard Medical School.

The first step was to find conditions better suited to the growth of Tritrichomonas than to the bacteria usually growing with it. This was accomplished by anaerobic cultivation in litre flasks of serum (Loeffler's dehydrated) saline medium run through Berkefeld filters. The contents of 10 litres were passed through filter paper and then concentrated by centrifugation. This gave 10 to 15 cc of Tritrichomonas having the appearance of heavy cream. Sixteen sterile 50 cc. pointed centrifuge tubes were filled with sterile culture medium near the freezing point, Tritrichomonas layered on surface centrifugalized 15 minutes at step 6 of rheostat, fluid drawn off, new tubes filled, Tritrichomonas layered on surface, centrifugalized, and so on for 15 to 20 times when the ratio of Tritrichomonas to bacteria was about 50 to 1. Tritrichomonas without living bacteria was placed in over 500 tubes of media containing various dead bacteria in different concentrations. It never multipled. Finally, it was obtained in a pure culture of a bacterial organism which did not grow at all at 36°C. It grew well. This afforded many opportunities. Tritrichomonas could now be maintained indefinitely at room temperature and transferred to all kinds of media with and without bacteria, losing the room temperature growing bacteria after one or two subcultures at 36°C. It never multiplied in culture media containing: (1) dead bacteria; (2) dead bacteria in unheated filtrate of living bacteria; (3) dead bacteria in unheated filtrates of living bacteria and green grass; (4) kidney, brain, liver. Frogs, rabbits, guinea-pigs had countless billions injected into blood stream with no growth.

A Tritrichomonas of Man in Pure Cultures of Various Microorganisms; Its Failure to Grow with Some and the Measurement of Its Growth and Division Rate with Others. L. R. Cleveland, Harvard Medical School.

After having obtained Tritrichomonas in a pure culture of bacteria which do not grow at all at 36°C. (see abstract above), but which grow well at 30°C. and

lower, it was then possible to determine the ability of this organism to grow in pure cultures of bacteria, molds, yeasts, and spirochetes in various media. It never multiplied with yeasts, molds, spirochetes, and certain bacteria *(Pseudomonas aeruginosa, Alcaligines fecalis);* with some bacteria *(Escherichia communior, Erbethella dysenteriae, E. typhi, Vibrio comma,* etc.) growth was very poor and cultures seldom ran for more than two to three transfers; with other bacteria *(Serratia marcescens, Staphylococcus citreus, S. tetragenus, Proteus vulgaris, Sarcina lutea,* etc.) growth was a little better; with others *(Salmonella enteritidis, Aerobacter aerogenes, Encapsulatus pneumoniae, Bacillus cereus, B. megatherium, B. subtilis, B. niger,* etc.) growth was good, with the last five bacteria Tritrichomonas became as numerous as blood flagellates do when grown on N. N. N. medium and formed agglomeration rosettes. When grown with *E. pneumoniae* at 36°C. the peak of reproduction occurred on the third or fourth day and no organisms were present after the sixth day; but when grown with the aerobic spore-forming bacteria the peak was reached much later (with *B. cereus* it was not reached until about the fifteenth day, while the numbers were not far below the peak after the sixth day). Growth with various bacteria will be shown in tabular form. There is no explanation as to why growth is much better with some bacteria than with others.

Preliminary Report on Observations on the Development of Ova of Pig and Human Ascaris under Natural Conditions, and Studies of Factors Influencing Development. F. C. and E. L. Caldwell, Field Research Laboratory, International Health Division, Rockefeller Foundation.

In search of an explanation of the distribution of Ascaris in the South Atlantic States, the development of ova of the pig and human Ascaris under natural conditions was observed throughout the year 1926-27 in South Alabama. Studies were made of the effect of soils, drying, heat, sunlight, and moisture under both cultural and natural conditions as determined by differential counts. Soils per se seem to have little influence in the epidemiology of Ascariasis. Ova in pig feces develop more rapidly and at a lower and also at higher temperatures than in human feces. Drying is the greatest lethal factor, ova in direct sunlight in high summer heat (98° F.) disintegrating in 3½ to 15 hours; under higher temperatures in incubators less rapidly; and so slowly in dry shade as to permit the development of ova to the infective stage in proportions varying with conditions. Trichuris ova disintegrate more rapidly than Ascaris. In deep shade under natural conditions in summer, ova in fecal "plants" develop in from 10 to 15 days to the motile stage. Kept moist in direct sunlight, development of ova is retarded; by the end of 30 days the majority of ova of human type have disintegrated without development, while the majority of pig ova contain motile embryos. Under conditions of artificial heat (40°-50°C.) ova of human feces kept moist fail to develop in 15 days and a large proportion disintegrate; ova of pig Ascaris develop slowly. When transferred to natural conditions, ova of the human type fail to develop in an additional 15 days while 85% of the pig Ascaris ova contain motile embryos. The moisture requirement of Ascaris ova, though definite, is low, development proceeding more rapidly with little moisture. Under saturation conditions, development is greatly retarded but not stopped.

The Structure and Development of Corallobothrium. Hiram E. Essex, University of Illinois.

Two new species of cestode which have been designated as *Corallobothrium giganteum* n. sp. and *C. fimbriatum* n. sp., are parasitic in *Ictalurus punctatus, Ameiurus melas, A. nebulosus,* and *Leptops olivarius.* Seventy per cent of the 130 *L. punctatus* examined harbored one or both species of cestode. The examinations were made from April to December, 1926. The adult parasites occurred only from spring to fall but plerocercoids were present throughout the year. The

life cycle of each parasite was studied experimentally during the summer of 1926. Infection with the larvae of *C. gigantcum* was produced by feeding the eggs of that species to *Cyclops serrulatus* and *C. prasinus*. Positive results were obtained by feeding the eggs of *C. fimbriatum* to *Cyclops bicuspidatus* and *C. serrulatus*. Oncospheres of *C. fimbriatum* were observed in the body-cavity of the Cyclops 4 hours after the feeding of the eggs. In each species development proceeded very rapidly. The body gradually elongated. A cercomer or bladder-appendage appeared at the posterior end. The anterior end then invaginated and the cercomer disappeared. This, the procercoid stage, was reached in *C. gigantcum* 11 to 12 days after feeding of the eggs while C. fimbriatum larvae required from 12 to 14 days to complete their development, in the Cyclops. Cyclops infected with the procercoides of C. fimbriatum were fed to *Notropis blennius*. The larvae were recovered from the body-cavity of the minnows 3 days after feeding. Young *L. punctatus* from 2½ to 3 inches long examined in September, 1927, harbored mature *C. fimbriatum*. Thus there is evidence that infection with this species may occur directly through the agency of the Cyclops or through a second intermediate host such as *N. blennius*.

The Cuticula of the Nemathelminthes. Justus F. Müller, University of Illinois.

The cuticulae of *Gordius robustus, Macracanthorhynchus hirudinaceus,* and *Ascaris lumbricoides* have been studied chemically. The cuticulae of Gordius and Macracanthorhynchus are chemically homogeneous. That of Ascaris can be separated into two separate chemical components, corresponding to the outer thin layer, or true cuticula, and the inner series of fibrous layers which constitute the major bulk of the cuticula. Each of these components was analyzed separately. The cuticulae of Acanthocephala and Gordius, and the two components of the cuticula of Ascaris constitute four distinct chemical compounds, all of which are proteins of albuminoid character. None of them shows any relation to chitin. The albuminoids of the cuticulae of Ascaris and Gordius are fairly similar in composition, while Acanthocephala cuticula is widely removed in its properties and composition from the rest. This is interesting in view of various views in regard to the relationships of this group. I can not agree with Magath that the cuticula of Ascaris is cornein as this statement rests upon an incorrect translation of the original German Reichard. The name *cornein* was given by Valenciennes in 1855 to the skeletal substance of the horny corals. This substance bears no anatomical homology or chemical relationship to the cuticula of Ascaris. The cuticula of Ascaris has now been correctly analyzed for the first time, since previous workers have analyzed this complex organ as a whole without first separating it into its elementary component parts.

A Study of the Immunity of Mosquitoes to Bird Malaria. Clay G. Huff, University of Georgia.

Studies upon the infectivity of Plasmodia of birds for mosquitoes for the purpose of testing as many as possible of the Culicine mosquitoes to ascertain which of them are vectors and for searching for the explanation of the non-susceptibility of certain species, have led to the following conclusions in regard to the immunity of mosquitoes to plasmodial infections. An immunity was found to exist in a certain percentage of the individuals of a species known to be vectors of the parasite in question. Using *Culex pipiens* as the species susceptible to, and *Aedes sollicitans* as the species non-susceptible to infection with *Plasmodium cathemerium*, it was found (1) that the length of life of the asexual forms in each mosquito was the same, (2) that digestion, in general, progressed at about the same rate in the stomachs of each species, and (3) that ookinetes appeared in each after the same interval of time. It would seem then that the immunity and susceptibility of these mosquitoes are to be explained in some way other than by the existence of different powers or rates of digestion in the two species.

Controlled Transmission of Plasmodium vivax *by* Anopheles punctipennis. J. H. St. John, Medical Corps, U. S. Army.

These observations were made possible by the use of *Anopheles punctipennis* in transmitting *Plasmodium vivax* to cases of syphilis of the central nervous system.

1. Case infections by *Anopheles punctipennis* were successful in 6 of 9 attempts.

2. Infection of different lots of mosquitoes varied in percent positive from 0 to 57%. The percentage results were determined by dissections, which, because of unavoidable loss of mosquitoes by death during the incubation period of the parasite within the mosquito, are admittedly inaccurate. Seven of 54 mosquitoes from several lots contained either oöcysts or sporozoites, giving an average positive percent of 12.9.

3. Two lots of mosquitoes were infected by feeding on two cases of mosquito induced malaria as early as four days after the onset of the clinical symptoms.

4. In one case, quinine in small doses did not stimulate the production of gametocytes.

5. The incubation period of mosquito-transmitted malaria ranged from 14 to 31 days.

6. The parasite curve in tertian malaria indicates that there is a "saturation" point as to the number present in the peripheral circulation at any stage of the infection. This point is reached in the majority of the infections followed for ten days, or more, when the parasites numbered approximately 1 to 2.5 leucocytes.

7. Comparison clinically of mosquito-induced tertian malaria and that produced by direct blood transfer disclosed little differences in the "reaction" of the patient to the parasite.

The Screw Worm Problem. F. C. Bishopp, Bureau of Entomology, U. S. Dept. of Agriculture.

An account is given of the screw worm as a parasite of man and animals especially in the Southwestern United States. Some of the habits and reactions of the screw worm are discussed and the steps outlined which may be taken to reduce the losses from this insect.

An Analysis of Variability in Hook Measurements in the Acanthocephala. H. J. Van Cleave, University of Illinois.

Proboscis hooks of the Acanthocephala have long been considered as providing characters for differentiating species. Lühe has given an analysis of variability in number and arrangement of the hooks of one species. To date there has been no critical consideration of factors responsible for variability in measurements. A series of measurements of hooks in the terminal circle of *Neoechinorhynchus cylindratus* gave values ranging from 53 to 100μ in length. Such variability is not a true measure of size differences for two groups of factors are involved, as follows: (a) actual size differences, (b) difference in measurements where no difference in size exists. Under this second foreshortening is the chief factor. In *N. cylindratus*, proboscis hooks in the same circle on the same individual if viewed in perfect profile give no indication of size differences. Shape of the hooks, orientation with reference to the axis of the body and to the optical plane produce conditions that create false impressions of size differences. Though hook size is not absolutely fixed for the species, actual size differences are much less prominent than range of measurements would indicate.

The Economic Importance of Veterinary Parasitology. Maurice C. Hall, Bureau of Animal Industry, U. S. Department of Agriculture.

On theoretical grounds, the limitations in the application of sanitation to livestock make parasitism of livestock a highly important topic, since domesticated animals, especially pasture animals, soil their table, the pasture, with the manure

bearing the worm eggs and larvae with which infection is renewed or spread to other susceptible animals, and their hairy or woolly hides make ideal playgrounds for external parasites. In practice the domesticated animal which is without parasites is a rare animal, seldom seen except in the case of those kept for years in the heart of a big city. Livestock, from horses and cows to chickens and pigeons, is attacked internally and externally by parasites representing hundreds of species, the horse alone having about 250 such parasites listed. The evidence indicates that for the most part these parasites are increasing in numbers and importance and are extending their distribution in the United States. The factors favoring the parasites are (1) the change from range conditions to farm conditions and (2) a remarkable transportation system by means of which animals may transplant their parasites to places a thousand miles apart. Man is controlling or defeating a handful of parasite species, and losing ground to all the others. The basic research on which control measures must be based has not even been attempted for most of these parasites. Present losses from parasites in America run into hundreds of millions of dollars annually, and this loss, which cannot be afforded now, may in a century mean a food shortage for coming generations.

Vitamin D and Resistance of Chickens to Parasitism. James E. Ackert and L. A. Spindler, Kansas State Agricultural College.

Five experiments involving 148 chickens were carried out to determine the effect of a lack of vitamin D upon the resistance of chickens to intestinal parasitism. Embryonated eggs of the fowl nematode, *Ascaridia lineata* (Schneider), were fed to young pure bred white leghorn chickens, which were not given vitamin D. An equal number of control chickens, from the same hatch, which were supplied with vitamin D were also parasitized in the same manner. The criterion for judging whether or not the lack of vitamin D lessened the resistance of these chickens to parasitism was the number and size of the worms that were able to remain in the minus D chicks compared with those in the control chickens. The results of the experiments lead the writers to conclude that the lack of vitamin D does not lower the resistance of young chickens to this intestinal roundworm during a period of three weeks of parasitism. The experiments, however, seemed to indicate that young chickens given vitamin D are more resistant to the effects of the parasites than are birds not supplied with this vitamin.

A Nodular Disease of the Intestine of the Carabao Due to a Species of the Trichostrongyle Genus Cooperia. Benjamin Schwartz, Bureau of Animal Industry, U. S. Department of Agriculture.

A portion of the small intestine of a carabao calf (*Bubalus bubalus*), forwarded to the present writer from the Philippine Islands for diagnosis, revealed the presence of numerous macroscopically visible nodules in the mucosa, each nodule containing a single sexually mature male or female nematode belonging to an undescribed species of the trichostrongyle genus Cooperia. The apex of each nodule is somewhat depressed and shows a small opening which represents the channel of communication between the parasite and the lumen of the intestine. The worms lie deeply in the nodule and are very much twisted apparently owing to the technic of fixation. Aside from the interest which attaches to these nematodes owing to the fact that they represent a new species and have a rather unusual location, they are apparently of considerable pathogenic importance because the animal in which the lesions were discovered died as a result of inanition. The ante-mortem symptoms were those of severe emaciation and debility. This nodular disease differs from other known nodular diseases of ruminants in that the incitant in the present case is a sexually mature worm belonging to the Trichostrongylidae, whereas nodular disease of cattle, sheep and other ruminants in the United States and elsewhere is due to the invasion of the intestinal mucosa by larvae of Strongylidae belonging to the genus Oesophagostomum.

Experimental Definitive Hosts of Thelazia callipaeda. Ernest Carroll Faust, Peking Union Medical College.

The finding of a natural infection of *Thelazia callipaeda* in the conjunctival sac of a laboratory rabbit in Peking in 1926 suggested the possibility that various mammals might serve as the definitive hosts of this nematode. Accordingly adult worms taken from conjunctival membranes of four naturally infected dogs were directly introduced into the conjunctival sac of several mammals, with the following results.

The data show that the dog and the rabbit were both suitable hosts for these worms; that the monkey was a suitable host up to the time of its death; that the worms failed to live in one cat after one week, did not survive in two cats after the first month but remained alive in another for six months; and that the goat and sheep appear to be entirely unsuitable hosts for the worms. In no case was there an increase in the number of the worms from that originally implanted, while in a few cases the number of worms recovered was slightly less. Evidence is furnished showing that there is considerable latitude in the choice of hosts but that certain animals are apparently unsuited for this nematode. The records of natural infection of the dog, man and rabbit with *Thelazia callipaeda* closely parallel the series of optimum hosts determined by the experimental method.

Human Infection with Larval Thelazia. Ernest Carroll Faust and Chen-hsiang Hu, Peking Union Medical College.

A foreigner, living in Chengtu, Szechuan, West China, where dogs commonly harbor the eye-worm, *Thelazia callipaeda,* was referred to the eye clinic of the Peking Union Medical College for removal of a wart-like papilloma of the skin on the lower right eyelid, not far from the internal canthus. The papilloma had been present for many years but during the previous fifteen months had slowly increased in size. On account of its itching the patient frequently rubbed the papilloma, which performance resulted in the development of a scablike incrustation around the lesion. During this latter period the patient gave a history of having frequently fondled a pet dog which had a severe irritation of both eyes. The papilloma was removed under local anesthesia and on section showed a chronic inflammation, with metozoan parasites in the epithelial folds and in the sebaceous glands. On examination the parasites proved to be larval nematodes, fairly well advanced in development. Several worms were present in each fold or pocket; in each case the heads were attached to the host tissue while the caudal extremities were free. The serrated pseudo-segmented character of the integument and the triradiate type of capsular thickening on either side of the mouth were unmistakable proof that the worms belonged to the genus Thelazia. Epidemiological evidence strongly favored the view that the species was *T. callipaeda.* This stage of the worm has never before been observed. Its location in a papilloma of the human skin is believed to be atypical but suggests that the larvae may normally secure attachment to inflamed tissues and continue their development. If this tissue is the conjunctival membrane the worms may proceed to maturity.

Intestinal Parasites in the Eastern Carolinas. W. G. Gamble, Jr., Medical College, Charleston, S. C.

By fecal examinations a survey was made covering three thousand (3,000) hospital cases admitted to a private hospital in northeastern South Carolina over a period of two years. All cases were considered, colored as well as white. Cases were separated according to counties and districts. Two or more examinations were made and both concentration and plain smear methods were used. The warm stage was not employed routinely. On account of the fact that these were almost all private cases getting fresh specimens was quite a problem. The survey included parasites as well as ova. Total infection was found to be 19%, all of which were recognized pathogenic parasites or ova. Treatment was instituted. The percentage of reinfection or incomplete treatment was 2%.

The Relation of Parasitism to Wild Life Conservation. E. C. O'Roke, University of California.

Food and water, shelter or cover, and protection from enemies are the factors usually considered in planning for the conservation of wild life. The important but little understood questions of parasitism are beginning to command the attention of naturalists and biologists. Points of contact for the transmission of parasites are areas where the interests of domestic animals and wild animals overlap. It is probable that many of the perplexing problems of game bird introductions are those dealing with parasites and parasitic diseases. Many of the parasites of water fowl are intimately related to the aquatic fauna of the ponds and marshes where birds feed. The parasitologist can render valuable service to game commissions and wild life institutes through his knowledge of the ways of parasites

BOOK REVIEWS

Studies on Clonorchis sinensis (Cobbold). By Ernest Carroll Faust and Oo-Key Khaw assisted by Yao Ke-Fang and Chao Yung-An. With a Consideration of the Molluscan Hosts of Clonorchis sinensis in Japan, China and Southeastern Asia and Other Species of Molluscs Closely Related to Them. By Bryant Walker, 284 pp., 33 figs. American Journal of Hygiene, Monographic Series, No. 8, March 1927.

To the previous studies on Schistosoma, Fasciolopsis, and hookworm in China published in the Monographic Series, is now added this exhaustive treatise on another important human parasite from the Far East. In a way this species has commanded wide attention recently in this country because of the publicity it has acquired thru quarantine regulations. Accordingly this monograph is most timely with its wealth of accurate information concerning the parasite and the disease it produces. In China the Clonorchis problem is of outstanding importance altho it has not been studied before this, and the results of earlier investigations in Japan are partly unknown and partly discredited among Western workers. Faust and Khaw have studied the problem intensively and their results as here presented enlarge our knowledge materially and bring final evidence on many disputed points.

The monograph opens with a historical résumé of the literature on this species followed by a list of references to earlier articles. The second chapter covers biological and morphological studies on *Clonorchis sinensis* and embraces more than one third of the entire work. It is replete with interesting new data. Among these, special mention should be made of Doctor Khaw's studies on the egg and its relations to its environment. Each stage in development is described with great exactitude and a firm basis is laid for the differential diagnosis of the ova which are very likely to be confused with those of other species. Statistical data are given on egg-production and its variation, on the influence of temperature, of saline waters, of night soil mixtures, etc. After describing the infection of the snail intermediate host and the cercaria, the author takes up the encysted stage in the fish and the subsequent transfer to the mammalian or final host. Substantially all the fresh water fish of the Sino-Japanese area are found to harbor the cysts of this parasite, altho cysts of other species were also present and not always readily distinguished. A consideration of the adult worm closes this section. The authors have made a very large number of observations and experiments on different phases and relations in this life history.

The third section deals with the distribution of the parasite, particularly in China, tho some data are given for certain other areas. The extent and relative abundance of the parasite is represented on shaded charts. The data show clearly that Clonorchis is very common in reservoir hosts (dogs and cats) in the Yangtze valley whereas human infection which is common in other parts is extremely rare in central China and entirely absent in North China: On the other hand hospital records from Canton demonstrate that some 30% of the population there is infected. Inquiry showed positively that the habit of eating raw fish though unsuspected was wide spread in the Canton district and that the fishes commonly used for food were generally infected. However reservoir hosts were only scantily infected in this region and are of little importance in spreading the infection in the Canton delta. This and one other prefecture, also in Kwangtung Province, constitute the most important centers of Clonorchis infection of the human population in China. While poorly cooked, or even dried and salted fresh-water fish may account for a small part of the infection, the practise of eating raw fresh-water fish is responsible for the large majority of cases. There are no important reservoir hosts in this province to carry Clonorchis

infection, and this stands in sharp contrast with conditions in Central China where such hosts are mainly responsible, or in North China where they are solely responsible.

The fourth section of the monograph deals with experimental therapy in Clonorchis infections, and the fifth with the prevention of the disease in man. Many carefully conducted experiments are discussed and the results analyzed critically. At the close of their work the authors write:

From the viewpoint of human medicine the extensive areas of Central and North China, where Clonorchis infection is common in fishes and in reservoir hosts, but is almost never contracted by man, constitute a most striking practical test of man's ability to live side by side with the potential infection without contracting it. The only known reason for the absence of human infection in these extensive areas is that the population of Central and North China is accustomed to cook all fish consumed. This difference in the habits of the Central and North China population from that of South China is probably a racial one, since the peoples of the south-coast provinces have closer kinship to the fish-eating Malay populations further south than do the true Chinese type of Central China or the mongoloid type in the north. Furthermore, absence of endemic human clonorchiasis in Central and North China constitutes a very practical demonstration in refutation of the alleged danger of infected Chinese introducing clonorchiasis into the United States. Such an argument would have been reasonable only in case the American population was itself generally disposed to eat uncooked fish. On the whole we feel that altogether too much stress has been placed on the "contagiousness" and "loathsomeness" of Clonorchis infection, which, as far as China is concerned, is confined to a relatively small area, and, furthermore, seldom incapacitates the human host.

The monograph includes as its sixth chapter a contribution by Bryant Walker on the molluscan intermediate hosts and closely related species. Doctor Walker's outstanding reputation in this field and his thorough treatment of all phases of the question make his section of great value for students of parasitology. The work closes with an exhaustive bibliography arranged both alphabetically and chronologically. It it particularly valuable since it includes all the Japanese references which are partially and often only vaguely known abroad and unfortunately are frequently erroneously cited. These Japanese papers are fully treated in the text of the monograph.

PREVENTIVE MEDICINE AND HYGIENE. By M. J. ROSENAU. (5th Ed.) 1458 pp. 157 figs. D. Appleton & Company, New York and London.

The early editions of this well-known work are so frequently consulted and so highly esteemed that workers in all associated fields have looked forward with eagerness for the appearance of a new edition and their hopes are thoroughly justified. The amount of labor bestowed upon the new volume is evidently large for, as the author states, altho no outstanding discoveries have characterized the interval of 5 years since the publication of the last edition, yet so many important contributions have been made to nearly every phase of the subject that the book has been substantially rewritten and entirely reset. This statement of the situation is peculiarly true of those phases that deal with animal parasites and parasitic diseases and their relation to preventive medicine and hygiene. The author has made good use of the abundant contributions of recent years in this field. Notably amoebic disentery, and balantidial disentery have been comprehensively treated in the light of recent studies. Among other new material on diseases caused by micro-organisms, the accounts of sleeping sickness and tularemia may be mentioned. One of the few slips noted was in the case of the latter disease as no reference was made to the important demonstration of the existence of tularemia in Illinois.

An extended section has been introduced on flukes and the diseases caused by them. While the parasitologist will welcome this emphasis upon a topic of

evident importance, yet it seems probable that in other editions this section will be somewhat increased and certainly modified by virtue of further evidence that is accumulating; part of this has been published too recently for the author to have included it in this work. The extensive treatment given insect borne diseases will appeal to all parasitologists and the accompanying tables give in brief form with conspicuous clarity the important facts concerned therewith. Students of animal parasitology will find the volume not only interesting and stimulating from the general standpoint but of specific value in their work.

PRÉCIS DE MÉDICINE COLONIALE. By CH. JOYEUX, 831 pp. 139 figs. Masson & Co., Paris

The appearance of this work by one of the leading French parasitologists is of especial interest because of the important role in tropical diseases played by animal parasites and because of the authorship of the work. Dr. Joyeux is peculiarly favored by his contacts with the famous Laboratory of Parasitology in the Faculty of Medicine at Paris. That city is conspicuous also for the numerous conferences on colonial affairs which are held there and by frequent visits from colonial medical officers. The author maintains also constant relations with field workers in the French colonies and other tropical regions, to many of whom he expresses individual thanks in the preface to the work under consideration. The book is written in form to be immediately useful to the colonial practitioner, the chapters resting upon a clinical classification of the diseases. Under each malady material is carefully organized and clearly set forth under a series of distinct headings. The sections dealing with geographic distribution and pathogenic agents which interest primarily the parasitologist have been very carefully worked up and present in brief form a picture of conditions that is thoroughly up to date. It is perhaps unfortunate that the author could not have used more illustrations of the causal organisms and the transmitting agents which are so important on the one hand in diagnosis and on the other hand in disease prevention. However the illustrations which are given should be commended for their natural appearance and freedom from the excessive diagrammatic accuracy which though generally sought serves to impart an unfamiliar appearance to the objects since the features presented are those that appear only after long study and are not demonstrable even on critical examination of fresh material. On the whole the author has produced a most attractive work and one that will be extremely useful in the hands of workers in the field.

PRACTICAL BACTERIOLOGY, BLOOD WORK AND ANIMAL PARASITOLOGY. By E. R. STITT, (8th Ed.), 837 pp. 211 figs. P. Blakiston's Son & Co., Philadelphia, Pa.

The textbook by Dr. Stitt has always commanded the approval of teachers and its popularity is evidenced by the numerous editions which have been issued. This, the eighth, has been revised and considerably enlarged. Changes in the section which deals with animal parasites have been extensive and bring the work thoroughly up to date. Indeed Part III, Animal Parasitology, which now covers more than 200 pages, constitutes in itself one of the best brief texts on the subject which is available. Unlike some medical works, adequate attention has been given to those matters which can only be decided by the professional zoologist and parasitologist. Dr. Stitt's advisors in this field have been of the best and he has shown good judgment in utilizing their advice. For diagnostic purposes the figures of flukes and tapeworms might have been improved by adding alongside of the illustration an index of the magnification. The illustrations of the ova of these parasites are seriously marred by the relatively excessive size of the type used in labelling the figures. These are unquestionably minor criticisms of a fine piece of work. However, it is difficult to understand why any scientific

man in this day should fill up a text of scientific character with English measurements and that too often expressed in vulgar fractions.

City Health Administration. By Carl E. McCombs, 524 pp. The Macmillan Company, New York City.

The attractive presentation of what is unquestionably an oft neglected though fundamentally important municipal problem is to be highly commended from the standpoint of effective and thoroughgoing treatment of the subject. The sections on Disease Prevention, Food Inspection, Sanitary Inspection, etc., include discussions of the relations to these subjects of parasitic diseases. Insect vectors and their relation to disease transmission with the means of guarding against their multiplication and functioning in this undesirable rôle are also the subject of comment. While some might wish that certain of these factors had received greater emphasis as befitting the results newly achieved in individual places, yet the presentation here deserves praise because these topics are too often passed over with no more than the briefest mention.

In the *Handbuch der Biologischen Arbeitsmethoden* (Vol. IX. Part 1), by Emil Abderhalden (Urban & Schwarzenberg, Berlin), W. A. Collier, of Buenos Aires, has written two brief but useful essays on the methods of investigating parasitic worms and on culture methods for handling those organisms. The author was unduly conservative in referring to American literature and has made no mention of pioneer work like that of Metcalf (Trans. Amer. Mic. Soc., 24: 89-102, Nov., 1903) for raising nematodes in cultures which was early and successfully developed in this country.

The appearance of the first numbers of the *Bulletin of the Antivenin Institute of America* commands attention because of the high standards it sets. This publication which is sure to be of interest to all workers in the field of medical zoology covers a field to which relatively little attention has been paid previously in America. The personnel of the Institute and the character of the first two numbers justify in highest degree the establishment of the new periodical.

Researches in Polynesia and Melanesia (Parts I-IV, Medical Entomology) by Patrick A. Buston and G. H. E. Hopkins, recently published as the first of the Memoir Series of the London School of Hygiene and Tropical Medicine, may by virtue of its general title escape the attention of parasitologists. While some general topics are considered, the bulk of the memoir is devoted to medical entomology and is worthy of careful attention. Special note should be made of the experimental study of egg laying, of the larvae and other features in the life history and bionomics of Aedes.

Researches on the Parasitology of Plague (Ceylon Journal of Science, D, 1: 279-448; May 16, 1927) by L. Fabian Hirst presents a most thorough discussion of the rat fleas, their geographical distribution and the relative role of various species in the spread of plague in nature. Especial attention should be called to the final section which discusses parasitology and plague prevention.

In *Vorlesungen Über Theoretische Mikrobiologie* by Julius Springer, Berlin. August Rippel has given a very interesting and complete tho condensed discussion of the biological processes of the bacteria with only occasional references to the parasitic protozoa. In view of the effectiveness with which the author has handled the discussion it is to be regretted that he did not expand the work sufficiently to include also other animal micro-organisms.

The Laboratory of Parasitology of the Faculty of Medicine at Sao Paulo, Brazil, has undertaken the publication of a new serial entitled *Boletim Biologico.* The numbers which have thus far appeared give promise that the publication will be of marked value to students of parasitology. Protozoa, nematodes, trematodes and insect carriers are among the topics discussed in systematic, morphological and biological articles. The publication is well printed and illustrated.

The *Thirteenth Annual Report of the International Health Board* which covers the year 1926 is full of interesting and valuable material for the student of

animal parasites. No one studying or teaching in the field should neglect to examine this work which is too rich in detail to receive more extended consideration here.

A comparative study of *Dengue* by J. F. Siler, Milron W. Hall and A. Parker Hitchens has just been published as Monograph 20 of the Bureau of Science at Manila. The history of disease, its relation to mosquitoes and methods of control are treated exhaustively and furnish most valuable information for the student of parasitology.

CORRIGENDA

In the September number the article entitled Notes on the Caryophyllaeidae by George W. Hunter, III, was by accident printed without the footnote in the text. This note is as follows:

* Contribution from the Zoological Laboratory of the University of Illinois under the direction of Henry B. Ward, No. 306.